Concepts of Genetics

TWELFTH EDITION

Klug • Cummings • Spencer • Palladino • Killian

Brief Contents

CONCEPTS OF GENETICS

TWELFTH EDITION
GLOBAL EDITION

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Michael R. Cummings is a Research Professor in the Department of Biological, Chemical, and Physical Sciences at Illinois Institute of Technology, Chicago, Illinois. For more than 25 years, he was a faculty member in the Department of Biological Sciences and in the Department of Molecular Genetics at the University of Illinois at Chicago. He has also served on the faculties of Northwestern University and Florida State University. He received his B.A. from St. Mary's College in Winona, Minnesota, and his M.S. and Ph.D. from Northwestern University in Evanston, Illinois. In addition to this text, he has written textbooks in human genetics and general biology. His research interests center on the molecular organization and physical mapping of the heterochromatic regions of human acrocentric chromosomes. At the undergraduate level, he teaches courses in molecular genetics, human genetics, and general biology, and has received numerous awards for teaching excellence given by university faculty, student organizations, and graduating seniors. When not teaching or writing, Dr. Cummings can often be found far offshore fishing for the one that got away.

Charlotte A. Spencer is a retired Associate Professor from the Department of Oncology at the University of Alberta in Edmonton, Alberta, Canada. She has also served as a faculty member in the Department of Biochemistry at the University of Alberta. She received her B.Sc. in Microbiology from the University of British Columbia and her Ph.D. in Genetics from the University of Alberta, followed by postdoctoral training at the Fred Hutchinson Cancer Research Center in Seattle, Washington. Her research interests involve the regulation of RNA polymerase II transcription in cancer cells, cells infected with DNA viruses, and cells traversing the mitotic phase of the cell cycle. She has taught undergraduate and graduate courses in biochemistry, genetics, molecular biology, and oncology. She has also written booklets in the Prentice Hall Exploring Biology series. When not writing and editing contributions to genetics textbooks, Dr. Spencer works on her hazelnut farm and enjoys the peace and quiet of a remote Island off the west coast of British Columbia.

Michael A. Palladino is Vice Provost for Graduate Studies, former Dean of the School of Science, and Professor of Biology at Monmouth University in West Long Branch, New Jersey. He received his B.S. degree in Biology from The College of New Jersey and his Ph.D. in Anatomy and Cell Biology from the University of Virginia. For more than 15 years he directed a laboratory of undergraduate student researchers supported by external funding from the National Institutes of Health, biopharma companies, and other agencies. He and his undergraduates studied molecular mechanisms involved in innate immunity of mammalian male reproductive organs and genes involved in oxygen homeostasis and ischemic injury of the testis. He has taught a wide range of courses including genetics, biotechnology, endocrinology, and cell and molecular biology. He has received several awards for research and teaching, including the 2009 Young Andrologist Award of the American Society of Andrology, the 2005 Distinguished Teacher Award from Monmouth University, and the 2005 Caring Heart Award from the New Jersey Association for Biomedical Research. He is co-author of the undergraduate textbook *Introduction to Biotechnology*. He was Series Editor for the Benjamin Cummings *Special Topics in Biology* booklet series, and author of the first booklet in the series, *Understanding the Human Genome Project*. When away from the university or authoring textbooks, Dr. Palladino can often be found watching or playing soccer or attempting to catch most any species of fish in freshwater or saltwater.

Darrell J. Killian is an Associate Professor and current Chair of the Department of Molecular Biology at Colorado College in Colorado Springs, Colorado. He received his B.A. degree in Molecular Biology and Biochemistry from Wesleyan University in Middletown, Connecticut, prior to working as a Research Technician in Molecular Genetics at Rockefeller University in New York, New York. He earned his Ph.D. in Developmental Genetics from New York University in New York, New York, and received his postdoctoral training at the University of Colorado—Boulder in the Department of Molecular, Cellular, and Developmental Biology. Prior to joining Colorado College, he was an Assistant Professor of Biology at the College of New Jersey in Ewing, New Jersey. His research focuses on the genetic regulation of animal development, and he has received funding from the National Institutes of Health and the National Science Foundation. Currently, he and his undergraduate research assistants are investigating the molecular genetic regulation of nervous system development using *C. elegans* and *Drosophila* as model systems. He teaches undergraduate courses in genetics, molecular and cellular biology, stem cell biology, and developmental neurobiology. When away from the classroom and research lab, Dr. Killian can often be found on two wheels exploring trails in the Pike and San Isabel National Forests.

Dedication

We dedicate this edition to our long-time colleague and friend Harry Nickla, who sadly passed away in 2017. With decades of experience teaching Genetics to students at Creighton University, Harry's contribution to our texts included authorship of the *Student Handbook and Solutions Manual* (for the US edition) and the test bank, as well as devising most of the Extra Spicy problems at the end of each chapter. He was also a source of advice during the planning session for each new edition, and during our many revisions. We always appreciated his professional insights, friendship, and conviviality. We were lucky to have him as part of our team, and we miss him greatly.

WSK, MRC, CAS, MAP, and DJK

Brief Contents

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Explore Cutting-Edge Topics

Concepts of Genetics **emphasizes the fundamental ideas of genetics, while exploring modern techniques and applications of genetic analysis. This best-selling text continues to provide understandable explanations of complex, analytical topics and recognizes the importance of teaching students how to become effective problem solvers.**

Six **Special Topics in Modern Genetics** mini-chapters concisely explore cutting-edge, engaging, and relevant topics.

- **NEW!** CRISPR-Cas and Genome Editing
- DNA Forensics
- Genomics and Precision Medicine
- Genetically Modified Foods
- Gene Therapy
- **NEW!** Advances in Neurogenetics: The Study of Huntington Disease

Special Topic chapters include Review and Discussion questions, which are also assignable in Mastering Genetics.

CRISPR-Cas and Genome Editing

Genetic research is often a slow incremental process that may extend our understanding of a concept or improve the efficiency of a genetic technology. More rarely, discoveries advance the field in sudden and profound ways. For example, studies in the early 1980s led to the discovery of catalytic RNAs, which transformed how geneticists think about RNA. Around the same time, the development of the polymerase chain reaction (PCR) provided a revolutionary tool for geneticists and other scientists. Rapid and targeted DNA amplification is now indispensable to genetic research and medical science. Given this context, one can appreciate how rare and significant a discovery would be that both illuminates a novel genetic concept as well as yields a new technology for genetics research and application. CRISPR-Cas is exactly that.

For over a century, scientists have studied the biological warfare between bacteria and the viruses that infect them. However, in 2007, experiments confirmed that bacteria have a completely novel defense mechanism against viruses known as CRISPR-Cas. This discovery completely changed the scope of our understanding of how bacteria and viruses combat one another, and coevolve. Moreover, the CRISPR-Cas system has now been adapted as an incredibly powerful tool for genome editing.

The ability to specifically and efficiently edit a genome has broad implications for research, biotechnology, and medicine. For decades, geneticists have used various strategies for genome editing with many successes, but also with limited efficiency and a significant investment of time and resources. CRISPR-Cas has been developed into an efficient, cost-effective molecular tool that can introduce precise and specific edits to a genome. It is not without its limitations, but it represents a technological leap, which we have not seen, arguably, since the innovation of PCR.

"CRISPR-Cas has been developed into an efficient, cost-effective molecular tool that can introduce precise and specific edits to a genome."

The discovery of CRISPR-Cas has impacted genetics and other related fields at an unprecedented pace (Figure ST 1.1). CRISPR-Cas is the focus of numerous patent applications and disputes, has been approved for use in clinical trials to treat disease, has been used to edit the genome of human embryos as a proof of concept for future medical applications, has instigated international discussions on its ethical use, and is most deserving of its own chapter in a genetics textbook.

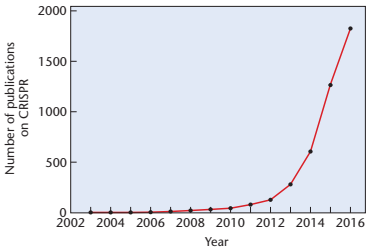

FIGURE ST 1.1 The number of publications returned in a search for "CRISPR" in PubMed by year.

ST 1.1 CRISPR-Cas Is an Adaptive Immune System in Prokaryotes

Bacteria and viruses (bacteriophages or phages) engage in constant biological warfare. Consequently, bacteria exhibit a diverse suite of defense mechanisms. For example, bacteria express endonucleases (restriction enzymes), which cleave specific DNA sequences. Such restriction enzymes destroy foreign bacteriophage DNA, while the bacterium protects its own DNA by methylating it. As you know (from Chapter 20), restriction enzymes have been adopted by molecular biologists for use in recombinant DNA technology. Bacteria can also defend against phage attack by blocking phage adsorption, blocking phage DNA insertion, and inducing suicide in infected cells to prevent the spread of infection to other cells. All of these defense mechanisms are considered **innate immunity** because they are not tailored to a specific pathogen.

Explore the Latest Updates

The 12th edition has been heavily updated throughout, including a reorganization and expansion of coverage of gene regulation in eukaryotes. This expansion reflects our growing knowledge of the critical roles RNA and epigenetics play in regulating gene activity.

NEW! Gene regulation in eukaryotes has been expanded into three chapters: transcriptional regulation (Ch. 17), posttranscriptional regulation (Ch. 18), and epigenetic regulation (Ch. 19).

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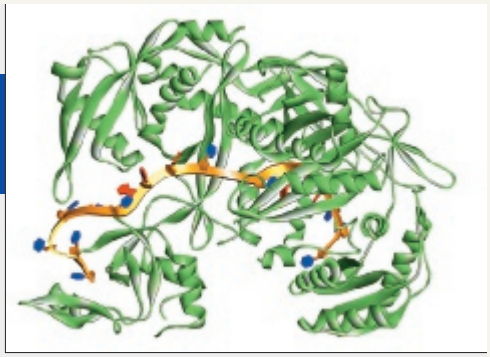

Posttranscriptional Regulation in Eukaryotes

Crystal structure of human Argonaute2 protein interacting with "guide" RNA. Argonaute2 plays an important role in mediating a posttranscriptional RNA-induced silencing pathway.

CHAPTER CONCEPTS

- Following transcription, there are several mechanisms that regulate gene expression, referred to as posttranscriptional regulation.
- Alternative splicing allows for a single gene to encode different protein isoforms with different functions.
- The interaction between *cis*-acting mRNA sequence elements and *trans*-acting RNA-binding proteins regulates mRNA stability, degradation, localization, and translation.
- Noncoding RNAs may regulate gene expression by targeting mRNAs for destruction or translational inhibition.
- Posttranslational modification of proteins can alter their activity or promote their degradation.

and the synthesis of a 3′ poly-A tail. Each of these steps can be regulated to control gene expression. After mature mRNAs are exported to the cytoplasm, they follow different paths: They may be localized to specific regions of the cell; they may be stabilized or degraded; or they may be translated robustly or stored for translation at a later time. Even after translation, protein activity, localization, and stability can be altered through covalent protein modifications. These and other eukaryotic posttranscriptional regulatory mechanisms are summarized in Figure 18.1.

Whereas the regulation of transcription depends on transcription factors and DNA regulatory elements (see Chapter 17), many posttranscriptional mechanisms involve RNA-level regulation. Moreover, posttranscriptional regulation is not only centered *on* RNA, but, in some cases, is regulated *by* RNA. Noncoding RNAs play important roles in the regulation of eukaryotic gene expression.

In this chapter, we will explore several important mechanisms and themes of eukaryotic posttranscriptional regulation. As you read on, keep in mind that while scientists have learned a great deal about how genes are regulated at the posttranscriptional level, there are still many unanswered questions for the curious student to ponder.

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Epigenetic Regulation of Gene Expression

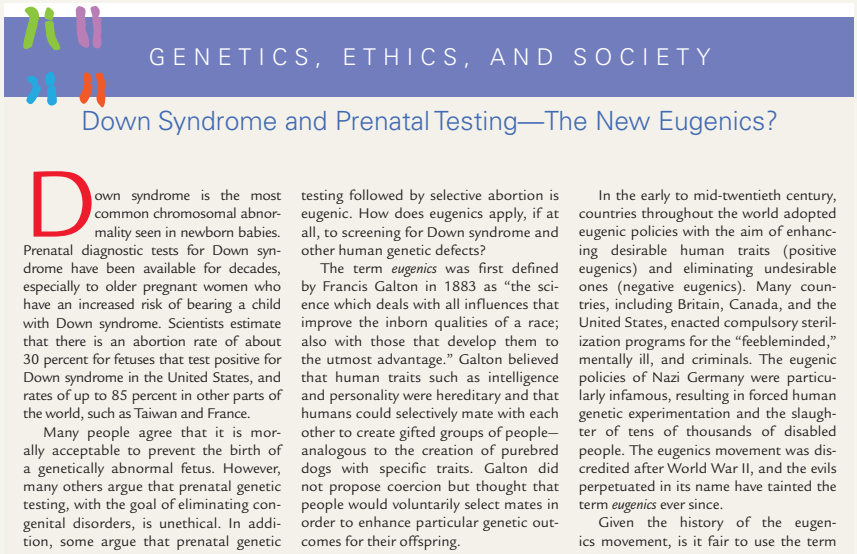
In toadflax, the shape of individual flowers changes from bilateral symmetry (photo on the left) to radial symmetry (photo on the right) in a naturally occurring, heritable gene silencing epimutation associated with the methylation of a single gene. There is no alteration of the DNA sequence at this locus.

CHAPTER CONCEPTS

NEW! A new chapter focuses on epigenetics, updating and expanding coverage that used to be in a Special Topics chapter.

and Ethical Considerations

With the rapid growth of our understanding of genetics and the ongoing introduction of powerful tools that can edit genes and genomes, it's important to encourage students to confront ethical issues and consider questions that arise in the study of genetics.



GENETICS, ETHICS, AND SOCIETY

Down Syndrome and Prenatal Testing—The New Eugenics?

Down syndrome is the most common chromosomal abnormality seen in newborn babies. Prenatal diagnostic tests for Down syndrome have been available for decades, especially to older pregnant women who have an increased risk of bearing a child with Down syndrome. Scientists estimate that there is an abortion rate of about 30 percent for fetuses that test positive for Down syndrome in the United States, and rates of up to 85 percent in other parts of the world, such as Taiwan and France.

Many people agree that it is morally acceptable to prevent the birth of a genetically abnormal fetus. However, many others argue that prenatal genetic testing, with the goal of eliminating congenital disorders, is unethical. In addition, some argue that prenatal genetic testing followed by selective abortion is eugenic. How does eugenics apply, if at all, to screening for Down syndrome and other human genetic defects?

The term *eugenics* was first defined by Francis Galton in 1883 as "the science which deals with all influences that improve the inborn qualities of a race; also with those that develop them to the utmost advantage." Galton believed that human traits such as intelligence and personality were hereditary and that humans could selectively mate with each other to create gifted groups of people—analogous to the creation of purebred dogs with specific traits. Galton did not propose coercion but thought that people would voluntarily select mates in order to enhance particular genetic outcomes for their offspring.

In the early to mid-twentieth century, countries throughout the world adopted eugenic policies with the aim of enhancing desirable human traits (positive eugenics) and eliminating undesirable ones (negative eugenics). Many countries, including Britain, Canada, and the United States, enacted compulsory sterilization programs for the "feebleminded," mentally ill, and criminals. The eugenic policies of Nazi Germany were particularly infamous, resulting in forced human genetic experimentation and the slaughter of tens of thousands of disabled people. The eugenics movement was discredited after World War II, and the evils perpetuated in its name have tainted the term *eugenics* ever since.

Given the history of the eugenics movement, is it fair to use the term

NEW! Genetics, Ethics, and Society essays appear in many chapters. Each one provides a synopsis of an ethical issue, related to chapter content, that impacts society today. Each includes a section called Your Turn, directing students to resources to help them explore the issue and answer questions.

NEW and REVISED! Case Studies conclude each chapter, introducing a short vignette of an everyday genetics-related situation and posing several discussion questions, including one focusing on ethics.

CASE STUDY Fish tales

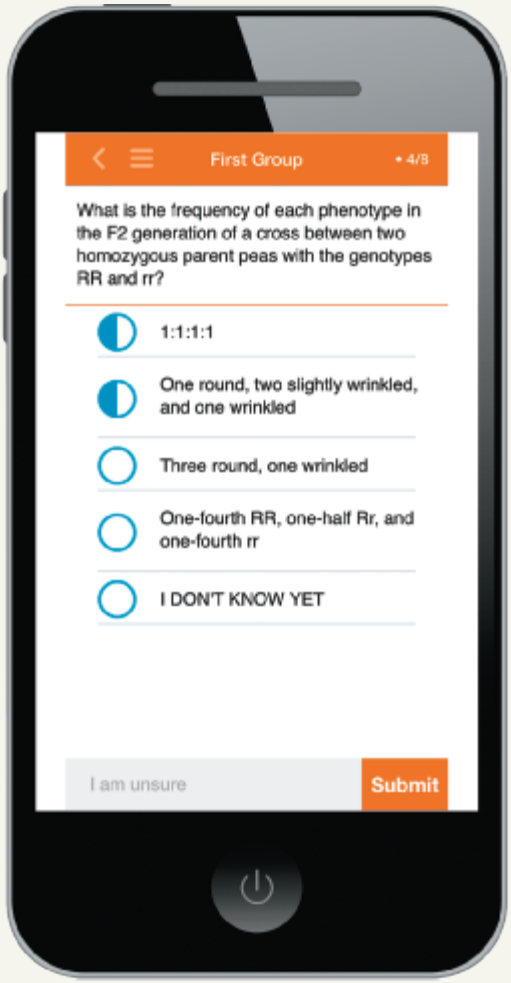
Controlling the overgrowth of invasive aquatic vegetation is a significant problem in the waterways of most U.S. states. Originally, herbicides and dredging were used for control, but in 1963, diploid Asian carp were introduced in Alabama and Arkansas. Unfortunately, through escapes and illegal introductions, the carp spread rapidly and became serious threats to aquatic ecosystems in 45 states. Beginning in 1983, many states began using triploid, sterile grass carp as an alternative, because of their inability to reproduce, their longevity, and their voracious appetite. On the other hand, this genetically modified exotic species, if not used properly, can reduce or eliminate desirable plants and outcompete native fish, causing more damage than good. The use of one exotic species to control other exotic species has had a problematic history across the globe, generating controversy and criticism. Newer methods for genetic modification of organisms to achieve specific outcomes will certainly become more common in the future and raise several interesting questions.

1. Why would the creation and use of a tetraploid carp species be unacceptable in the above situation?
2. If you were a state official in charge of a particular waterway, what questions would you ask before approving the use of a laboratory-produced, triploid species in this waterway?
3. What ethical responsibilities accompany the ecological and economic risks and benefits of releasing exotic species into the environment? Who pays the costs if ecosystems and food supplies are damaged?

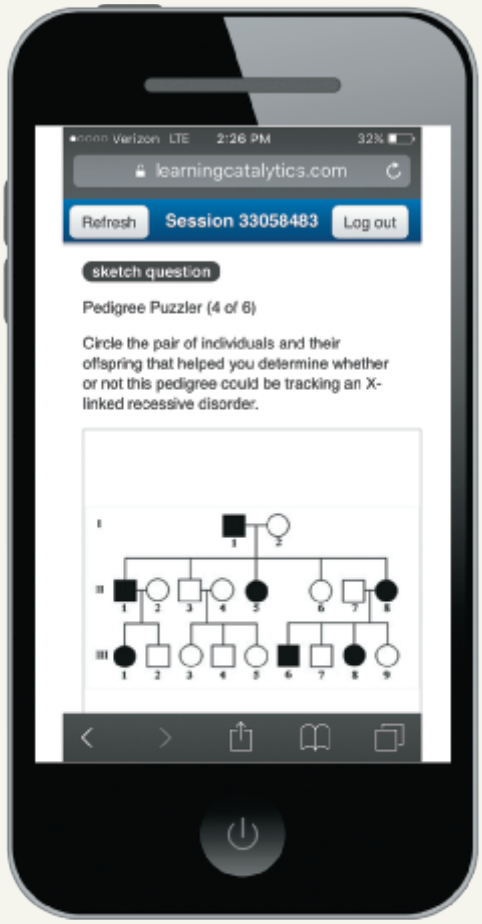
See Seastedt, T. R. (2015). Biological control of invasive plant species: A reassessment for the Anthropocene. *New Phytologist* 205:490–502.

Learn Genetics Concepts and Problem Solving

Mastering™ Genetics helps students master key genetics concepts while reinforcing problem-solving skills with hints and feedback specific to their misconceptions. Mastering Genetics includes content and tools for before, during, and after class. Learn more at www.pearson.com/mastering/genetics

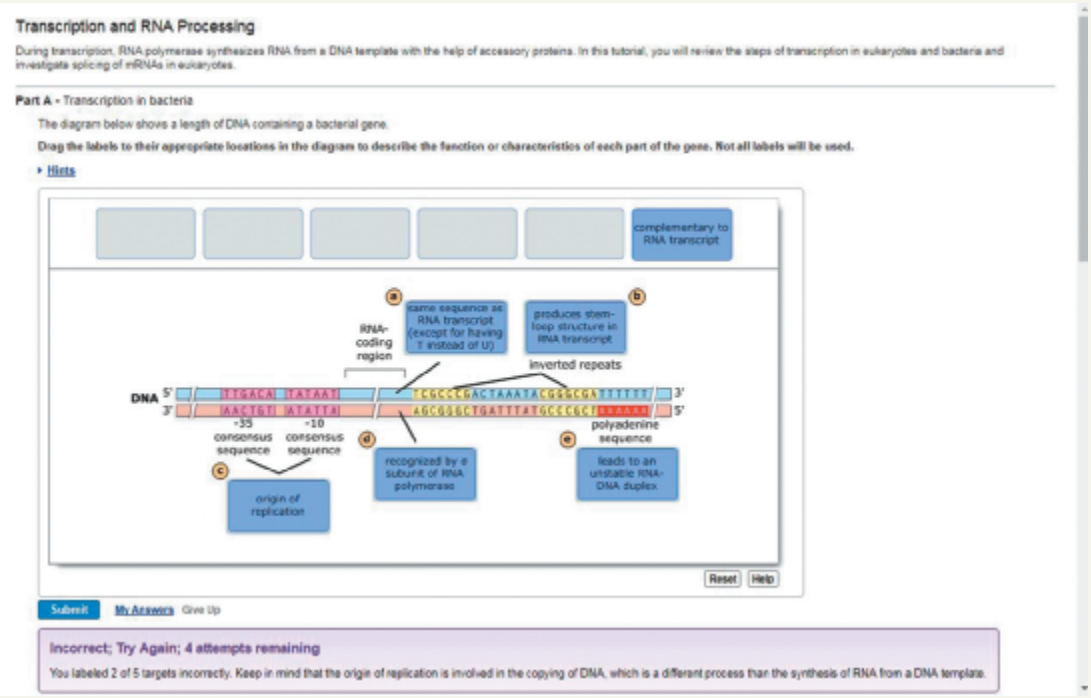


NEW! Dynamic Study Modules personalize each student's learning experience. Available for assignments or for self-study, these chapter-based modules help prepare students for in-class discussions, problem solving, or active learning. A mobile app is available for iOS and Android devices.



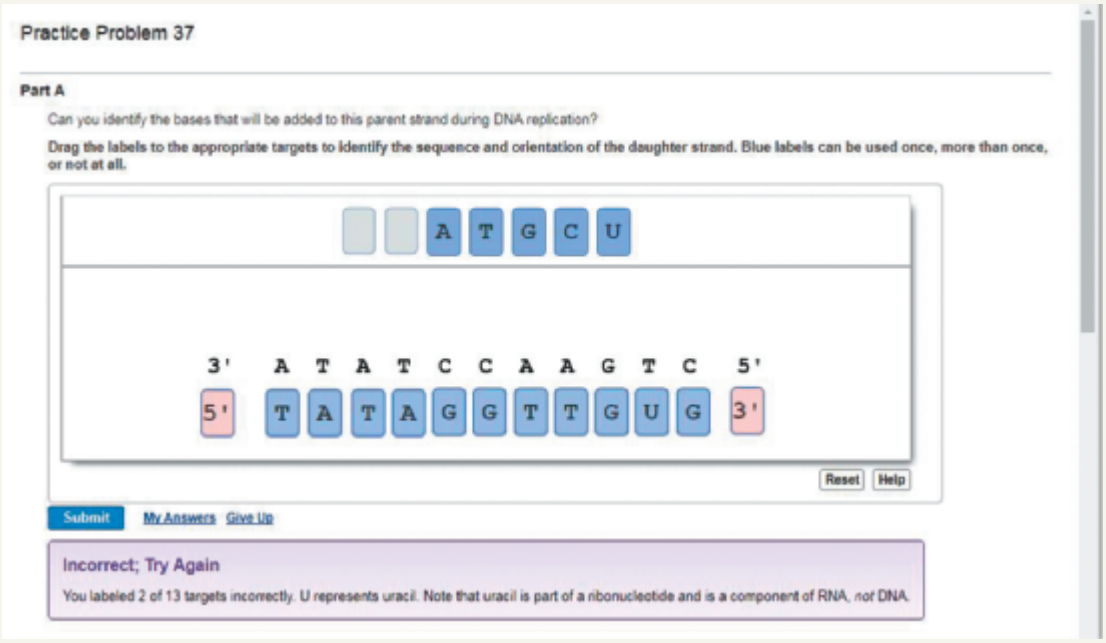
Learning Catalytics is a "bring your own device" (smartphone, tablet, or laptop) assessment and active classroom system that helps engage students. Instructors can create their own questions, draw from community content, or access Pearson's library of question clusters.

with Mastering Genetics



Tutorials and activities feature personalized wrong-answer feedback and hints that emulate the office-hour experience to guide student learning.

100 Practice Problems offer more opportunities to develop problem-solving skills. These questions appear only in Mastering Genetics and include targeted wrong-answer feedback to help students learn.



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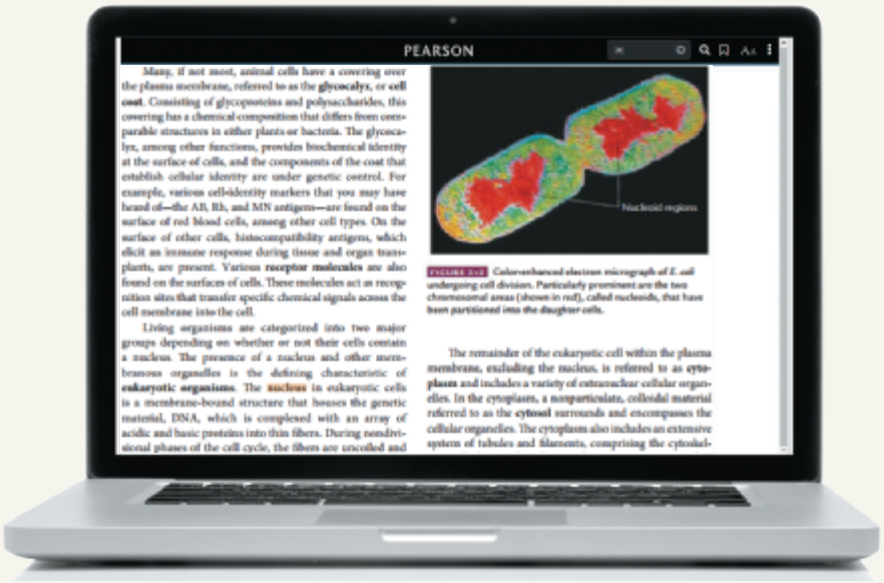

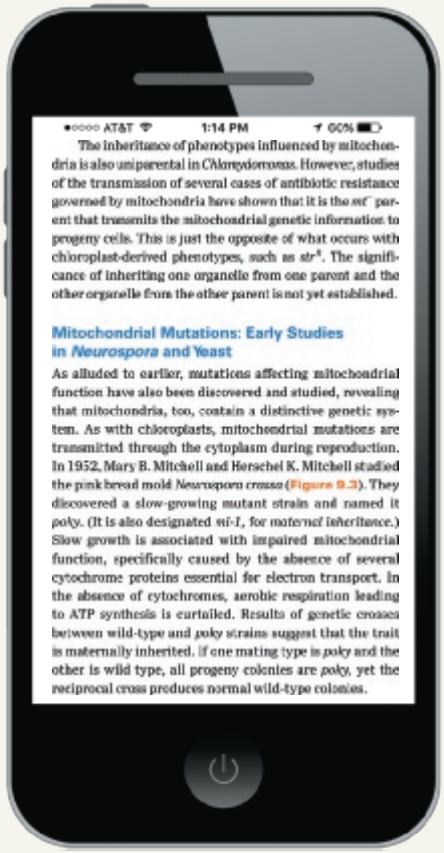

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Preface

It is essential that textbook authors step back and look with fresh eyes as each edition of their work is planned. In doing so, two main questions must be posed: (1) How has the body of information in their field—in this case, Genetics—grown and shifted since the last edition? (2) Which pedagogic innovations that are currently incorporated into the text should be maintained, modified, or deleted? The preparation of the 12th edition of *Concepts of Genetics*, a text well into its fourth decade of providing support for students studying in this field, has occasioned still another fresh look. And what we focused on in this new edition, in addition to the normal updating that is inevitably required, were three things:

1. **The importance of continuing to provide comprehensive coverage of important, emerging topics.**

   In this regard, we continue to include a unique approach in genetics textbooks that offers readers a set of abbreviated, highly focused chapters that we label **Special Topics in Modern Genetics**. In this edition, these provide unique, cohesive coverage of six important topics: *CRISPR-Cas and Genomic Editing*, *DNA Forensics*, *Genomics and Precision Medicine*, *Genetically Modified Foods*, *Gene Therapy*, and *Advances in Neurogenetics: The Study of Huntington Disease*. The initial and final chapters in this series are both new to this edition.

2. **The recognition of the vastly increased knowledge resulting from the study of gene regulation in eukaryotes.**

   To that end, the single chapter on this topic in previous editions has been expanded to three chapters: "Transcriptional Regulation in Eukaryotes" (Chapter 17), "Posttranscriptional Regulation in Eukaryotes" (Chapter 18), and "Epigenetic Regulation of Gene Expression" (Chapter 19). This extended coverage reflects many recent discoveries that reveal that RNA in many forms other than those that are essential to the process of transcription and translation (mRNA, tRNA, and rRNA) play critical roles in the regulation of eukaryotic gene activity. As well, it is now clear based on molecular studies related to epigenetics that this topic is best taught as an integral part of eukaryotic gene regulation. This new material provides the student exposure to modern coverage of a significant research topic.

3. **The importance of providing an increased emphasis on ethical considerations that genetics is bringing into everyday life.**

   Regarding this point, we have converted the essay feature *Genetics, Technology, and Society* to one with added emphasis on ethics and renamed it *Genetics, Ethics, and Society*. Approximately half the chapters have new or revised essays. In addition, the feature called *Case Study*, which appears near the end of all chapters, has been recast with an increased focus on ethics. Both of these features increase the opportunities for active and cooperative learning.

Goals

In the 12th edition of *Concepts of Genetics*, as in all past editions, we have five major overarching goals. Specifically, we have sought to:

- Emphasize the basic concepts of genetics.
- Write clearly and directly to students, providing understandable explanations of complex, analytical topics.
- Maintain our strong emphasis on and provide multiple approaches to problem solving.
- Propagate the rich history of genetics, which so beautifully illustrates how information is acquired during scientific investigation.
- Create inviting, engaging, and pedagogically useful full-color figures enhanced by equally helpful photographs to support concept development.

These goals collectively serve as the cornerstone of *Concepts of Genetics*. This pedagogic foundation allows the book to be used in courses with many different approaches and lecture formats.

Writing a textbook that achieves these goals and having the opportunity to continually improve on each new edition has been a labor of love for all of us. The creation of each of the twelve editions is a reflection not only of our passion for teaching genetics, but also of the constructive feedback and encouragement provided by adopters, reviewers, and our students over the past four decades.

New to This Edition

New to this edition are four chapters. Two are Special Topics in Modern Genetics entries entitled "CRISPR-Cas and Genome Editing" and "Advances in Neurogenetics: The Study of Huntington Disease." Both cover cutting-edge information and represent very recent breakthroughs in genetics. CRISPR, a genome-editing tool, is a straightforward technique that allows specific, highly accurate modification of DNA sequences within genes and is thus a powerful tool in the world of genetic research and gene therapy. In addition to this chapter, we call your attention to the introduction to Chapter 1 for an introduction to CRISPR and to also note that we have chosen this gene-editing system as the subject matter illustrated on the cover. Special Topics Chapter 6 illustrates the many of advances that have been made in the study of human neurogenetics. Huntington disease, a monogenic human disorder, has been subjected to analysis for over 40 years using every major approach and technique developed to study molecular genetics, and as such, exemplifies the growing body of information that has accrued regarding its causes, symptoms, and future treatment.

Additional new chapters arise from a major reorganization and expansion of our coverage of regulation of gene expression in eukaryotes, where we have split our previous coverage into three parts: transcriptional regulation (Chapter 17), posttranscriptional regulation (Chapter 18), and epigenetic regulation (Chapter 19). Chapter 18 includes much of the content previously contained in the Special Topics chapter *Emerging Roles of RNA* in the previous edition. Chapter 19, focused on epigenetics, is an expansion of the content previously contained in the *Epigenetics* Special Topics chapter from the previous edition.

Collectively, the addition of these four new chapters provides students and instructors with a much clearer, up-to-date presentation to these important aspects of genetics.

Continuing Pedagogic Features

We continue to include features that are distinct from, and go beyond, the text coverage, which encourage active and cooperative learning between students and the instructor.

- **Modern Approaches to Understanding Gene Function** This feature highlights how advances in genetic technology have led to our modern understanding of gene function. Appearing in many chapters, this feature prompts students to apply their analytical thinking skills, linking the experimental technology to the findings that enhance our understanding of gene function.
- **Genetics, Ethics, and Society** This feature provides a synopsis of an ethical issue related to a current finding in genetics that impacts directly on society today. It includes a section called *Your Turn*, which directs students to related resources of short readings and Web sites to support deeper investigation and discussion of the main topic of each essay.
- **Case Study** This feature, at the end of each chapter, introduces a short vignette of an everyday genetics-related situation, followed by several discussion questions. Use of the Case Study should prompt students to relate their newly acquired information in genetics to ethical issues that they may encounter away from the course.
- **Evolving Concept of the Gene** This short feature, integrated in appropriate chapters, highlights how scientists' understanding of the gene has changed over time. Since we cannot see genes, we must infer just what this unit of heredity is, based on experimental findings. By highlighting how scientists' conceptualization of the gene has advanced over time, we aim to help students appreciate the process of discovery that has led to an ever more sophisticated understanding of hereditary information.
- **How Do We Know Question** Found as the initial question in the *Problems and Discussion Questions* at the end of each chapter, this feature emphasizes the pedagogic value of studying how information is acquired in science. Students are asked to review numerous findings discussed in the chapter and to summarize the process of discovery that was involved.
- **Concept Question** This feature, found as the second question in the *Problems and Discussion Questions* at the end of each chapter, asks the student to review and comment on common aspects of the Chapter Concepts, listed at the beginning of each chapter. This feature places added emphasis on our pedagogic approach of conceptual learning.
- **Mastering Genetics** This robust online homework and assessment program guides students through complex topics in genetics, using in-depth tutorials that coach students to correct answers with hints and feedback specific to their misconceptions. New content for the 12th edition of *Concepts of Genetics* includes Dynamic Study Modules and interactive flash cards that help students master basic content so they can be more prepared for class and for solving genetics problems.

New and Updated Topics

We have revised each chapter in the text to present the most current, relevant findings in genetics. Here is a list of some of the most significant new and updated topics covered in this edition.

Chapter 1: Introduction to Genetics
- New introductory vignette that discusses the discovery and applications of the genome-editing CRISPR-Cas system
- Updated section "We Live in the Age of Genetics"

Chapter 5: Sex Determination and Sex Chromosomes
- Updated content on the XIST gene product as a long noncoding RNA
- New insights about a novel gene involved in temperature-sensitive differentiation of snapping turtles and lizards, as well as the impact of climate change on sex, sex reversal, and sex ratios

Chapter 9: Extranuclear Inheritance
- Updated information on mtDNA disorders and nuclear DNA mismatches

Chapter 11: DNA Replication and Recombination
- New coverage of the role of telomeres in disease, aging, and cancer
- New and expanded coverage of telomeres and chromosome stability, explaining how telomeres protect chromosome ends

Chapter 13: The Genetic Code and Transcription
- New coverage on transcription termination in bacteria
- New section entitled "Why Do Introns Exist?"
- Updated coverage on RNA editing

Chapter 14: Translation and Proteins
- New coverage of eukaryotic closed-loop translation, including a new figure
- Revised coverage of Beadle and Tatum's classic experiments
- Expanded coverage on the posttranslational modifications of proteins
- New coverage of the insights gleaned from the crystal structure of the human 80*S* ribosome

Chapter 15: Gene Mutation, DNA Repair, and Transposons
- New and revised coverage on transposons, focusing on the mechanisms of transposition by both retrotransposons and DNA transposons, as well as a discussion of how transposition creates mutations. Two new tables and five new figures are included
- Reorganization of the mutation classification section with table summaries
- New and expanded coverage of human germ-line and somatic mutation rates

Chapter 17: Transcriptional Regulation in Eukaryotes
- Revised chapter organization focuses specifically on transcriptional regulation
- Revised coverage of regulation of the *GAL* gene system in yeast with an updated figure
- New coverage on genetic boundary elements called insulators

Chapter 18: Posttranscriptional Regulation in Eukaryotes
- New chapter that greatly expands upon the previous coverage of posttranscriptional gene regulation in eukaryotes
- Revised and expanded coverage of alternative splicing and its relevance to human disease
- Expanded coverage on RNA stability and decay with a new figure
- Updated coverage of noncoding RNAs that regulate gene expression with a new figure
- Enriched coverage of ubiquitin-mediated protein degradation with a new figure

Chapter 19: Epigenetic Regulation of Gene Expression
- New chapter emphasizing the role of epigenetics in regulating gene expression, including coverage of cancer, transmission of epigenetic traits across generations, and epigenetics and behavior
- New coverage on the recently discovered phenomenon of monoallelic expression of autosomal genes
- Updated coverage of epigenome projects

Chapter 20: Recombinant DNA Technology
- Increased emphasis on the importance of whole-genome sequencing approaches
- New coverage of CRISPR-Cas as a gene editing approach, including a new figure
- Updated content on next-generation and third-generation sequencing

Chapter 21: Genomic Analysis
- Increased emphasis on the integration of genomic, bioinformatic, and proteomic approaches to analyzing genomes and understanding genome function

- A new section entitled "Genomic Analysis Before Modern Sequencing Methods," which briefly summarizes approaches to mapping and identifying genes prior to modern sequencing
- Reorganized and revised content on the Human Genome Project. Updated content on personal genome projects and new content on diploid genomes and mosaicism and the pangenome to emphasize human genetic variations
- New coverage of the Human Microbiome Project including a new figure displaying microbiome results of patients with different human disease conditions
- New coverage of *in situ* RNA sequencing

Chapter 22: Applications of Genetic Engineering and Biotechnology

- Updated content on biopharmaceutical products including newly approved recombinant proteins, DNA vaccine trials to immunize against Zika virus, genetically modified organisms, and gene drive in mosquitos to control the spread of Zika
- New coverage of genes essential for life and how synthetic genomics is being applied to elucidate them. Clarification of prognostic and diagnostic genetics tests and the relative value of each for genetic analysis
- New content on DNA and RNA sequencing
- New section entitled "Screening the Genome for Genes or Mutations You Want," which discusses how scientists can look at genetic variation that confers beneficial phenotypes
- New section entitled "Genetic Analysis by Personal Genomics Can Include Sequencing of DNA and RNA" that expands coverage of personal genome projects and new approaches for single-cell genetic analysis of DNA and RNA

Chapter 23: Developmental Genetics

- New section entitled "Epigenetic Regulation of Development"
- New coverage of DNA methylation and progressive restriction of developmental potential
- Expanded coverage of binary switch genes and regulatory networks

Chapter 24: Cancer Genetics

- Extended coverage of environmental agents that contribute to human cancers, including more information about both natural and human-made carcinogens
- New section entitled "Tobacco Smoke and Cancer" explaining how a well-studied carcinogen induces a wide range of genetic effects that may lead to mutations and cancer
- New section entitled "Cancer Therapies and Cancer Cell Biology," describing the mechanisms of chemotherapies and radiotherapies as they relate to cancer cell proliferation, DNA repair, and apoptosis

Chapter 25: Quantitative Genetics and Multifactorial Traits

- Updated coverage on quantitative trait loci (QTLs)
- Revised and expanded section entitled "eQTLs and Gene Expression"

Chapter 26: Population and Evolutionary Genetics

- New coverage on vertebrate evolution
- New coverage of phylogenetic trees
- Updated coverage on the origins of the human genome
- New section entitled "Genotype and Allele Frequency Changes"
- New coverage on pre- and post-zygotic isolating mechanisms

Special Topic Chapter 1: CRISPR-Cas and Genome Editing

- New chapter on a powerful genome editing tool called CRISPR-Cas
- Up-to-date coverage on CRISPR-Cas applications, the patenting of this technology, and the ethical concerns of human genome editing

Special Topic Chapter 2: DNA Forensics

- New section on the still controversial DNA phenotyping method, including new explanations of how law-enforcement agencies currently use this technology

Special Topic Chapter 3: Genomics and Precision Medicine

- New section entitled "Precision Oncology," including descriptions of two targeted cancer immunotherapies: adoptive cell transfer and engineered T-cell therapies
- Updated pharmacogenomics coverage, including a description of new trends in preemptive gene screening for pharmacogenomic variants as well as the pGEN4Kids program, a preemptive gene screening program that integrates DNA analysis data into patient electronic health records

Special Topic Chapter 4: Genetically Modified (GM) Foods

- New section entitled "Gene Editing and GM Foods" describing how scientists are using the new techniques of gene editing (including ZFN, TALENS, and CRISPR-Cas) to create GM food plants and animals,

and how these methods are changing the way in which GM foods are being regulated
- A new box entitled "The New CRISPR Mushroom" describing the development and regulatory approval of the first CRISPR-created GM food to be approved for human consumption

Special Topic Chapter 5: Gene Therapy
- Updated coverage of gene therapy trials currently underway
- Reordered chapter content to highlight emergence of CRISPR-Cas in a new section entitled "Gene Editing"
- Substantially expanded content on CRISPR-Cas including a brief summary of some of the most promising trials in humans and animals to date
- Incorporation of antisense RNA and RNA interference into a new section entitled "RNA-based Therapeutics," including updated trials involving spinal muscular atrophy
- Updated content on roles for stem cells in gene therapy
- New content on combining gene editing with immunotherapy
- New ethical discussions on CRISPR-Cas and germline and embryo editing

Special Topic Chapter 6: Advances in Neurogenetics: The Study of Huntington Disease (HD)
- New chapter that surveys the study of HD commencing around 1970 up to the current time
- Coverage of the genetic basis and expression of HD, the mapping and isolation of the gene responsible for the disorder, the mutant gene product, molecular and cellular alterations caused by the mutation, transgenic animal models of HD, cellular and molecular approaches to therapy, and a comparison of HD to other inherited neurodegenerative disorders

Strengths of This Edition

- **Organization** —We have continued to attend to the organization of material by arranging chapters within major sections to reflect changing trends in genetics. Of particular note is the expansion of our coverage of the regulation of gene expression in eukaryotes, now reorganized into three chapters at the end of Part Three. Additionally, Part Four continues to provide organized coverage of genomics into three carefully integrated chapters.
- **Active Learning** —A continuing goal of this book is to provide features within each chapter that small groups of students can use either in the classroom or as assignments outside of class. Pedagogic research continues to support the value and effectiveness of such active and cooperative learning experiences. To this end, there are four features that greatly strengthen this edition: *Case Study*; *Genetics, Ethics, and Society*; *Exploring Genomics*; and *Modern Approaches to Understanding Gene Function*. Whether instructors use these activities as active learning in the classroom or as assigned interactions outside of the classroom, the above features will stimulate the use of current pedagogic approaches during student learning. The activities help engage students, and the content of each feature ensures that they will become knowledgeable about cutting-edge topics in genetics.

Emphasis on Concepts

The title of our textbook—*Concepts of Genetics*—was purposefully chosen, reflecting our fundamental pedagogic approach to teaching and writing about genetics. However, the word "concept" is not as easy to define as one might think. Most simply put, we consider a concept to be *a cognitive unit of meaning—an abstract representation that encompasses a related set of scientifically derived findings and ideas*. Thus, a concept provides a broad mental image that, for example, might reflect a straightforward snapshot in your mind's eye of what constitutes a chromosome; a dynamic vision of the detailed processes of replication, transcription, and translation of genetic information; or just an abstract perception of varying modes of inheritance.

We think that creating such mental imagery is the very best way to teach science, in this case, genetics. Details that might be memorized, but soon forgotten, are instead subsumed within a conceptual framework that is easily retained and nearly impossible to forget. Such a framework may be expanded in content as new information is acquired and may interface with other concepts, providing a useful mechanism to integrate and better understand related processes and ideas. An extensive set of concepts may be devised and conveyed to eventually encompass and represent an entire discipline—and this is our goal in this genetics textbook.

To aid students in identifying the conceptual aspects of a major topic, each chapter begins with a section called *Chapter Concepts*, which identifies the most important topics about to be presented. Each chapter ends with a section called *Summary Points*, which enumerates the five to ten key points that have been discussed. And in the *How Do We Know*? question that starts each chapter's problem set, students are asked to connect concepts to experimental findings. This question is then followed by a *Concept Question*, which asks the student to review and comment on common aspects of the Chapter Concepts. Collectively, these features help to ensure that students engage in, become aware of, and understand the major conceptual issues as they confront the extensive vocabulary and the many important details of genetics. Carefully designed figures also support our conceptual approach throughout the book.

Emphasis on Problem Solving

As authors and teachers, we have always recognized the importance of enhancing students' problem-solving skills. Students need guidance and practice if they are to develop into strong analytical thinkers. To that end, we present a suite of features in every chapter to optimize opportunities for student growth in the important areas of problem solving and analytical thinking.

- **Now Solve This** Found several times within the text of each chapter, each entry provides a problem similar to ones found at the end of the chapter that is closely related to the current text discussion. In each case, a pedagogic hint is provided to offer insight and to aid in solving the problem.
- **Insights and Solutions** As an aid to the student in learning to solve problems, the *Problems and Discussion Questions* section of each chapter is preceded by what has become an extremely popular and successful section. *Insights and Solutions* poses problems or questions and provides detailed solutions and analytical insights as answers are provided. The questions and their solutions are designed to stress problem solving, quantitative analysis, analytical thinking, and experimental rationale. Collectively, these constitute the cornerstone of scientific inquiry and discovery.
- **Problems and Discussion Questions** Each chapter ends with an extensive collection of *Problems and Discussion Questions*. These include several levels of difficulty, with the most challenging (*Extra-Spicy Problems*) located at the end of each section. Often, Extra-Spicy Problems are derived from the literature of genetic research, with citations. Brief answers to all even-numbered problems are presented in Appendix B.
- **How Do We Know?** Appearing as the first entry in the *Problems and Discussion Questions* section, this question asks the student to identify and examine the experimental basis underlying important concepts and conclusions that have been presented in the chapter. Addressing these questions will aid the student in more fully understanding, rather than memorizing, the endpoint of each body of research. This feature is an extension of the learning approach in biology first formally described by John A. Moore in his 1999 book *Science as a Way of Knowing—The Foundation of Modern Biology*.
- **Mastering Genetics** Tutorials in Mastering Genetics help students strengthen their problem-solving skills while exploring challenging activities about key genetics content. In addition, end-of-chapter problems are also available for instructors to assign as online homework. Students will also be able to access materials in the Study Area that help them assess their understanding and prepare for exams.

For the Instructor

Mastering Genetics—http://www.masteringgenetics.com

Mastering Genetics engages and motivates students to learn and allows you to easily assign automatically graded activities. Tutorials provide students with personalized coaching and feedback. Using the gradebook, you can quickly monitor and display student results. Mastering Genetics easily captures data to demonstrate assessment outcomes. Resources include:

- New Dynamic Study Modules, which are interactive flashcards, provide students with multiple sets of questions with extensive feedback so they can test, learn, and retest until they achieve mastery of the textbook material. These can be assigned for credit or used for self-study, and they are powerful preclass activities that help prepare students for more involved content coverage or problem solving in class.
- In-depth tutorials that coach students with hints and feedback specific to their misconceptions
- An item library of thousands of assignable questions including end-of-chapter problems, reading quizzes, and test bank items.
- Over 100 Practice Problems are like end-of-chapter questions in scope and level of difficulty and are found only in Mastering Genetics. The bank of questions extends your options for assigning challenging problems. Each problem includes specific wrong answer feedback to help students learn from their mistakes and to guide them toward the correct answer.
- The eText provides a dynamic digital version of the textbook. Its features include student and instructor note-taking, highlighting, bookmarking, search, and glossary.
- A gradebook that provides you with quick results and easy-to-interpret insights into student performance.

Downloadable Instructor Resources

The instructor resources for the 12th edition offers adopters of the text convenient access to a comprehensive and innovative set of lecture presentation and teaching tools. Developed to meet the needs of veteran and newer instructors alike, these resources include:

- The JPEG files of all text line drawings with labels individually enhanced for optimal projection results (as well as unlabeled versions) and all text tables.
- Most of the text photos, including all photos with pedagogical significance, as JPEG files.
- The JPEG files of line drawings, photos, and tables preloaded into comprehensive PowerPoint® presentations for each chapter.
- A second set of PowerPoint® presentations consisting of a thorough lecture outline for each chapter augmented by key text illustrations.
- PowerPoint® presentations containing a comprehensive set of in-class clicker questions for each chapter.
- An impressive series of concise instructor animations adding depth and visual clarity to the most important topics and dynamic processes described in the text.
- In Word and PDF files, a complete set of the assessment materials and study questions and answers from the testbank. Files are also available in TestGen format.

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1

Introduction to Genetics

Newer model organisms in genetics include the roundworm, *Caenorhabditis elegans*; the zebrafish, *Danio rerio*; and the mustard plant, *Arabidopsis thaliana*.

CHAPTER CONCEPTS

- Genetics in the twenty-first century is built on a rich tradition of discovery and experimentation stretching from the ancient world through the nineteenth century to the present day.
- Transmission genetics is the general process by which traits controlled by genes are transmitted through gametes from generation to generation.
- Mutant strains can be used in genetic crosses to map the location and distance between genes on chromosomes.
- The Watson–Crick model of DNA structure explains how genetic information is stored and expressed. This discovery is the foundation of molecular genetics.
- Recombinant DNA technology revolutionized genetics, was the foundation for the Human Genome Project, and has generated new fields that combine genetics with information technology.
- Biotechnology provides genetically modified organisms and their products that are used across a wide range of fields including agriculture, medicine, and industry.
- Model organisms used in genetics research are now utilized in combination with recombinant DNA technology and genomics to study human diseases.
- Genetic technology is developing faster than the policies, laws, and conventions that govern its use.

One of the small pleasures of writing a genetics textbook is being able to occasionally introduce in the very first paragraph of the initial chapter a truly significant breakthrough in the discipline that hopefully will soon have a major, diverse impact on human lives. In this edition, we are fortunate to be able to discuss the discovery of **CRISPR-Cas**, a molecular complex found in bacteria that has the potential to revolutionize our ability to rewrite the DNA sequence of genes from any organism. As such, it represents the ultimate tool in genetic technology, whereby the genome of organisms, including humans, may be precisely edited. Such gene modification represents the ultimate application of the many advances in biotechnology made in the last 35 years, including the sequencing of the human genome.

Other systems have been developed, including **zinc-finger nucleases (ZFNs)** and **transcription activator-like effector nucleases (TALENs)**, that are now undergoing clinical trials for the treatment of human diseases, and which we will discuss later in the text. However, the CRISPR-Cas system is the most powerful and far-reaching method and is now the preferred approach in gene modification. This system allows researchers to edit genomes with greater accuracy, is easier to use, and is more versatile than the ZFN or TALEN systems. CRISPR-Cas molecules were initially discovered as a molecular complex that protects bacterial cells

from invasion by viruses. CRISPR (clustered regularly interspersed short palindromic repeats) designates an RNA molecule, which in the laboratory can be synthesized to match any DNA sequence of choice. CRISPR RNA has two ends: one recognizes and binds to a matching DNA sequence in the gene of interest, and the other binds to a CRISPR-associated (Cas) nuclease, or DNA-cutting enzyme. The most commonly used Cas nuclease is Cas9, but there are many other Cas nucleases, each of which has slightly different properties, contributing to the system's versatility. In laboratory experiments, CRISPR-Cas systems have already been used to repair mutations in cells derived from individuals with several genetic disorders, including cystic fibrosis, Huntington disease, beta-thalassemia, sickle cell disease, muscular dystrophy, and X-linked retinitis pigmentosa, which results in progressive vision loss. In the United States a clinical trial using CRISPR-Cas9 for genome editing in cancer therapy has been approved, and a second proposal for treating a genetic form of blindness is in preparation. A clinical trial using CRISPR-Cas9 for cancer therapy is already under way in China.

The application of this remarkable system goes far beyond research involving human genetic disorders. In organisms of all kinds, wherever genetic modification may improve on nature to the benefit of human existence and of our planet, the use of CRISPR-Cas will find many targets. For example, one research group was able to use this system to spread genes that prevent mosquitoes from carrying the parasite that causes malaria. Other researchers have proposed using CRISPR-Cas9 to engineer laboratory-grown human blood vessels and organs that do not express proteins that cause rejection of transplanted tissues and organs. The method has also been used to create disease-resistant strains of wheat and rice.

The power of this system, like any major technological advance, has already raised ethical concerns. For example, genetic modification of human germ cells or embryos would change the genetic information carried by future generations. These modifications may have unintended and significant negative consequences for our species. An international summit on human gene editing in December 2015 concluded that a global forum to address concerns about heritable modifications should be convened to formulate regulations that apply to all countries involved in CRISPR research.

CRISPR-Cas may turn out to be one of the most exciting genetic advances in decades. We will return later in the text to an extended discussion of its discovery, describe how it works, its many applications, and the ethical considerations that it raises (see Special Topic Chapter 1—CRISPR and Genomic Editing).

For now, we hope that this short introduction has stimulated your curiosity, interest, and enthusiasm for the study of genetics. The remainder of this chapter provides an overview of major concepts of genetics and a survey of the major turning points in the history of the discipline. Along the way, enjoy your studies, but take your responsibilities as a novice geneticist most seriously.

1.1 Genetics Has a Rich and Interesting History

We don't know when people first recognized the hereditary nature of certain traits, but archaeological evidence (e.g., pictorial representations, preserved bones and skulls, and dried seeds) documents the successful domestication of animals and the cultivation of plants thousands of years ago by the artificial selection of genetic variants from wild populations. Between 8000 and 1000 B.C., horses, camels, oxen, and wolves were domesticated, and selective breeding of these species soon followed. Cultivation of many plants, including maize, wheat, rice, and the date palm, began around 5000 B.C. Such evidence documents our ancestors' successful attempts to manipulate the genetic composition of species.

During the Golden Age of Greek culture, the writings of the Hippocratic School of Medicine (500–400 B.C.) and of the philosopher and naturalist Aristotle (384–322 B.C.) discussed heredity as it relates to humans. The Hippocratic treatise *On the Seed* argued that active "humors" in various parts of the body served as the bearers of hereditary traits. Drawn from various parts of the male body to the semen and passed on to offspring, these humors could be healthy or diseased, with the diseased humors accounting for the appearance of newborns with congenital disorders or deformities. It was also believed that these humors could be altered in individuals before they were passed on to offspring, explaining how newborns could "inherit" traits that their parents had "acquired" in response to their environment.

Aristotle extended Hippocrates' thinking and proposed that the male semen contained a "vital heat" with the capacity to produce offspring of the same "form" (i.e., basic structure and capacities) as the parent. Aristotle believed that this heat cooked and shaped the menstrual blood produced by the female, which was the "physical substance" that gave rise to an offspring. The embryo developed not because it already contained the parts of an adult in miniature form (as some Hippocratics had thought) but because of the shaping power of the vital heat. Although the ideas of Hippocrates and Aristotle sound primitive and naive today, we should recall that prior to the 1800s neither sperm nor eggs had been observed in mammals.

1600–1850: The Dawn of Modern Biology

Between about 300 B.C. and 1600 A.D., there were few significant new ideas about genetics. However, between 1600 and 1850, major strides provided insight into the biological basis of life. In the 1600s, William Harvey studied reproduction and development and proposed the theory of **epigenesis**, which states that an organism develops from the fertilized egg by a succession of developmental events that eventually transform the egg into an adult. The theory of epigenesis directly conflicted with the theory of **preformation**, which stated that the fertilized egg contains a complete miniature adult, called a **homunculus** (Figure 1.1). Around 1830, Matthias Schleiden and Theodor Schwann proposed the **cell theory**, stating that all organisms are composed of basic structural units called cells, which are derived from preexisting cells. The idea of **spontaneous generation**, the creation of living organisms from nonliving components, was disproved by Louis Pasteur later in the century, and living organisms were then considered to be derived from preexisting organisms and to consist of cells.

In the mid-1800s the revolutionary work of Charles Darwin and Gregor Mendel set the stage for the rapid development of genetics in the twentieth and twenty-first centuries.

Charles Darwin and Evolution

With this background, we turn to a brief discussion of the work of Charles Darwin, who published *The Origin of Species*, in 1859, describing his ideas about evolution.

FIGURE 1.1 Depiction of the *homunculus*, a sperm containing a miniature adult, perfect in proportion and fully formed.

Darwin's geological, geographical, and biological observations convinced him that existing species arose by descent with modification from ancestral species. Greatly influenced by his voyage on the HMS *Beagle* (1831–1836), Darwin's thinking led him to formulate the theory of **natural selection**, which presented an explanation of the mechanism of evolutionary change. Formulated and proposed independently by Alfred Russel Wallace, natural selection is based on the observation that populations tend to contain more offspring than the environment can support, leading to a struggle for survival among individuals. Those individuals with heritable traits that allow them to adapt to their environment are better able to survive and reproduce than those with less adaptive traits. Over a long period of time, advantageous variations, even very slight ones, will accumulate. If a population carrying these inherited variations becomes reproductively isolated, a new species may result.

Darwin, however, lacked an understanding of the genetic basis of variation and inheritance, a gap that left his theory open to reasonable criticism well into the twentieth century. Shortly after Darwin published his book, Gregor Johann Mendel published a paper in 1866 showing how traits were passed from generation to generation in pea plants and offering a general model of how traits are inherited. His research was little known until it was partially duplicated and brought to light by Carl Correns, Hugo de Vries, and Erich Tschermak around 1900.

By the early part of the twentieth century, it became clear that heredity and development were dependent on genetic information residing in genes contained in chromosomes, which were then contributed to each individual by gametes—the so-called *chromosomal theory of inheritance*. The gap in Darwin's theory was closed, and Mendel's research has continued to serve as the foundation of genetics.

1.2 Genetics Progressed from Mendel to DNA in Less Than a Century

Because genetic processes are fundamental to life itself, the science of genetics unifies biology and serves as its core. The starting point for this branch of science was a monastery garden in central Europe in the late 1850s.

Mendel's Work on Transmission of Traits

Gregor Mendel, an Augustinian monk, conducted a decade-long series of experiments using pea plants. He applied quantitative data analysis to his results and showed that traits are passed from parents to offspring in predictable ways.

He further concluded that each trait in the plant is controlled by a pair of factors (which we now call genes) and that during gamete formation (the formation of egg cells and sperm), members of a gene pair separate from each other. His work was published in 1866 but was largely unknown until it was cited in papers published by others around 1900. Once confirmed, Mendel's findings became recognized as explaining the transmission of traits in pea plants and all other higher organisms. His work forms the foundation for **genetics**, which is defined as the branch of biology concerned with the study of heredity and variation. Mendelian genetics will be discussed later in the text (see Chapters 3 and 4).

The Chromosome Theory of Inheritance: Uniting Mendel and Meiosis

Mendel did his experiments before the structure and role of chromosomes were known. About 20 years after his work was published, advances in microscopy allowed researchers to identify chromosomes (**Figure 1.2**) and establish that, in most eukaryotes, members of each species have a characteristic number of chromosomes called the **diploid number (2*n*)** in most of their cells. For example, humans have a diploid number of 46 (**Figure 1.3**). Chromosomes in diploid cells exist in pairs, called **homologous chromosomes**.

Researchers in the last decades of the nineteenth century also described chromosome behavior during two forms of cell division, **mitosis** and **meiosis**. In mitosis (**Figure 1.4**), chromosomes are copied and distributed so that each daughter cell receives a diploid set of chromosomes identical to those in the parental cell. Meiosis is associated with gamete formation. Cells produced by meiosis receive only one chromosome from each chromosome pair, and the resulting number of chromosomes is called the **haploid number (*n*)**. This reduction in chromosome number is essential if the offspring arising from the fusion of egg and sperm are to maintain the constant number of chromosomes characteristic of their parents and other members of their species.

Early in the twentieth century, Walter Sutton and Theodor Boveri independently noted that the behavior

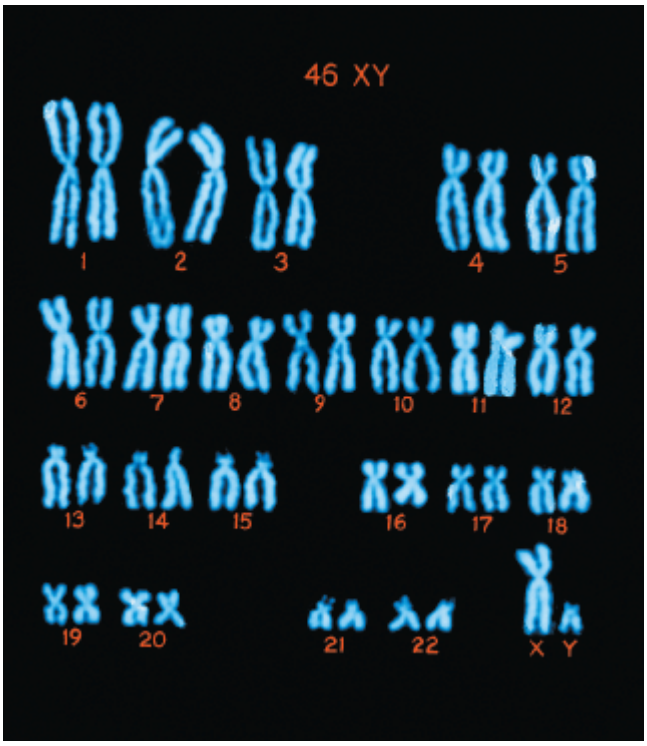


FIGURE 1.3 A colorized image of the human male chromosome set. Arranged in this way, the set is called a karyotype.

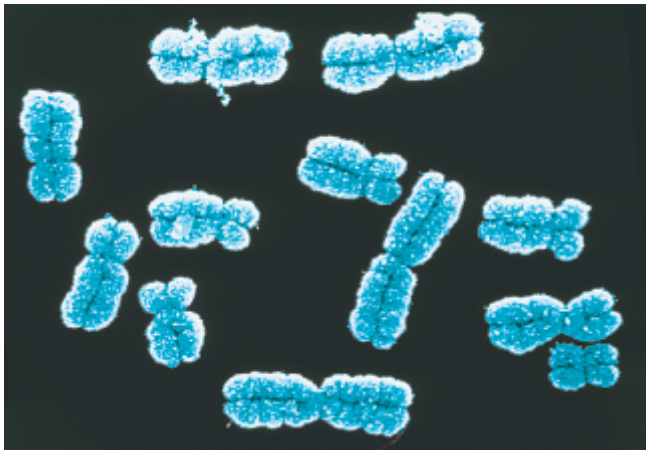


FIGURE 1.2 A colorized image of human chromosomes that have duplicated in preparation for cell division, as visualized using a scanning electron microscope.

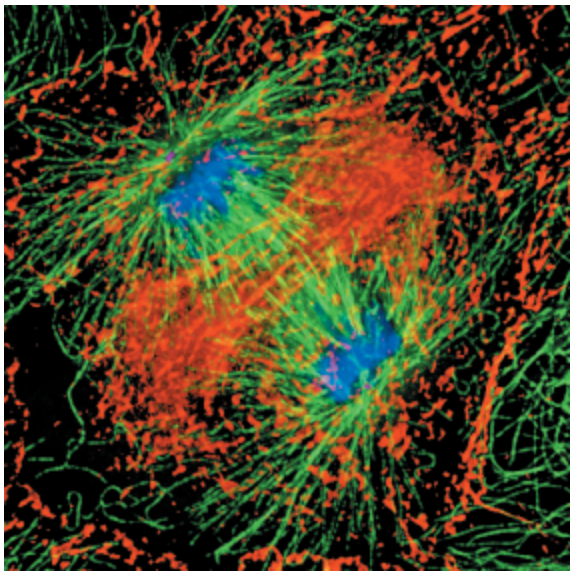


FIGURE 1.4 A late stage in mitosis after the chromosomes (stained blue) have separated.

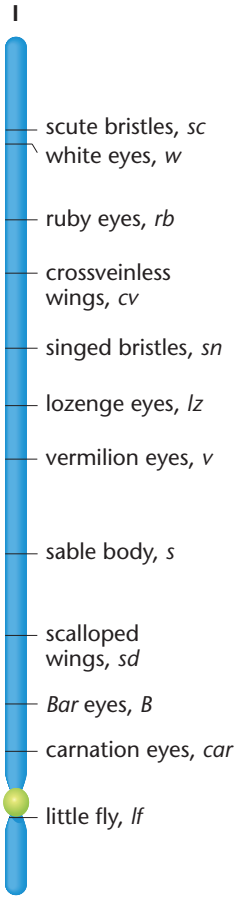


FIGURE 1.5 A drawing of chromosome I (the X chromosome, one of the sex-determining chromosomes) of *D. melanogaster*, showing the location of several genes. Chromosomes can contain hundreds of genes.

of chromosomes during meiosis is identical to the behavior of genes during gamete formation described by Mendel. For example, genes and chromosomes exist in pairs, and members of a gene pair and members of a chromosome pair separate from each other during gamete formation. Based on these and other parallels, Sutton and Boveri each proposed that genes are carried on chromosomes (**Figure 1.5**). They independently formulated the **chromosome theory of inheritance**, which states that inherited traits are controlled by genes residing on chromosomes faithfully transmitted through gametes, maintaining genetic continuity from generation to generation.

Genetic Variation

About the same time that the chromosome theory of inheritance was proposed, scientists began studying the inheritance of traits in the fruit fly, *Drosophila melanogaster*. Early in this work, a white-eyed fly (**Figure 1.6**) was discovered among normal (wild-type) red-eyed flies. This variation was produced by a **mutation** in one of the genes controlling

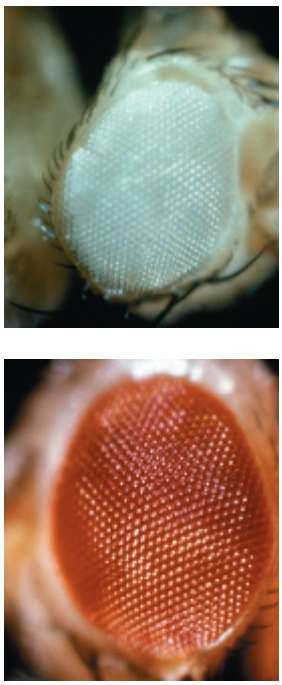


FIGURE 1.6 The white-eyed mutation in *D. melanogaster* (top) and the normal red eye color (bottom).

eye color. Mutations are defined as any heritable change in the DNA sequence and are the source of all genetic variation.

The white-eye variant discovered in *Drosophila* is an **allele** of a gene controlling eye color. Alleles are defined as alternative forms of a gene. Different alleles may produce differences in the observable features, or **phenotype**, of an organism. The set of alleles for a given trait carried by an organism is called the **genotype**. Using mutant genes as markers, geneticists can map the location of genes on chromosomes (Figure 1.5).

The Search for the Chemical Nature of Genes: DNA or Protein?

Work on white-eyed *Drosophila* showed that the mutant trait could be traced to a single chromosome, confirming the idea that genes are carried on chromosomes. Once this relationship was established, investigators turned their attention to identifying which chemical component of chromosomes carries genetic information. By the 1920s, scientists knew that proteins and DNA were the major chemical components of chromosomes. There are a large number of different proteins, and because of their universal distribution in the nucleus and cytoplasm, many researchers thought proteins were the carriers of genetic information.

In 1944, Oswald Avery, Colin MacLeod, and Maclyn McCarty, researchers at the Rockefeller Institute in New York, published experiments showing that DNA was the carrier

of genetic information in bacteria. This evidence, though clear-cut, failed to convince many influential scientists. Additional evidence for the role of DNA as a carrier of genetic information came from Hershey and Chase who worked with viruses. This evidence that DNA carries genetic information, along with other research over the next few years, provided solid proof that DNA, not protein, is the genetic material, setting the stage for work to establish the structure of DNA.

1.3 Discovery of the Double Helix Launched the Era of Molecular Genetics

Once it was accepted that DNA carries genetic information, efforts were focused on deciphering the structure of the DNA molecule and the mechanism by which information stored in it produces a phenotype.

The Structure of DNA and RNA

One of the great discoveries of the twentieth century was made in 1953 by James Watson and Francis Crick, who described the structure of DNA. DNA is a long, ladder-like macromolecule that twists to form a double helix (**Figure 1.7**). Each linear strand of the helix is made up of subunits called **nucleotides**. In DNA, there are four different nucleotides, each of which contains a nitrogenous base, abbreviated A (adenine), G (guanine), T (thymine), or C (cytosine). These four bases, in various sequence combinations, ultimately encode genetic information. The two strands of DNA are exact complements of one another, so that the rungs of the ladder in the double helix always consist of A=T and G=C base pairs. Along with Maurice Wilkins, Watson and Crick were awarded a Nobel Prize in 1962 for their work on the structure of DNA. We will discuss the structure of DNA later in the text (see Chapter 9).

Another nucleic acid, RNA, is chemically similar to DNA but contains a different sugar (ribose rather than deoxyribose) in its nucleotides and contains the nitrogenous base uracil in place of thymine. RNA, however, is generally a single-stranded molecule.

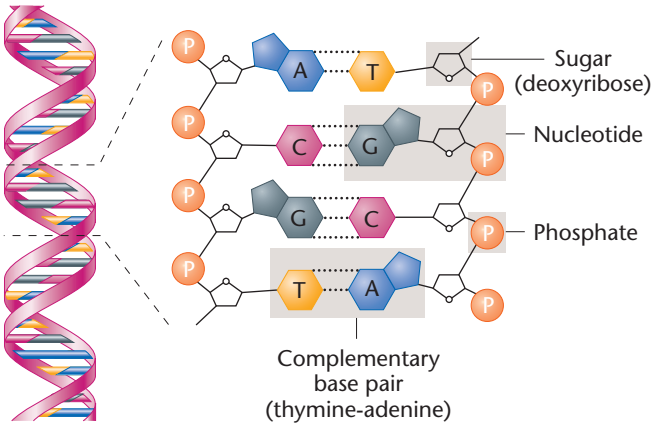


FIGURE 1.7 Summary of the structure of DNA, illustrating the arrangement of the double helix (on the left) and the chemical components making up each strand (on the right). The dotted lines on the right represent weak chemical bonds, called hydrogen bonds, which hold together the two strands of the DNA helix.

Gene Expression: From DNA to Phenotype

The genetic information encoded in the order of nucleotides in DNA is expressed in a series of steps that results in the formation of a functional gene product. In the majority of cases, this product is a protein. In eukaryotic cells, the process leading to protein production begins in the nucleus with **transcription**, in which the nucleotide sequence in one strand of DNA is used to construct a complementary RNA sequence (top part of **Figure 1.8**). Once an RNA molecule is produced, it moves to the cytoplasm, where the RNA—called **messenger RNA**, or **mRNA** for short—binds to a **ribosome**. The synthesis of proteins under the direction of mRNA is called **translation** (center part of Figure 1.8). The information encoded in mRNA (called the **genetic code**) consists of a linear series of nucleotide triplets. Each triplet, called a **codon**, is complementary to the information stored

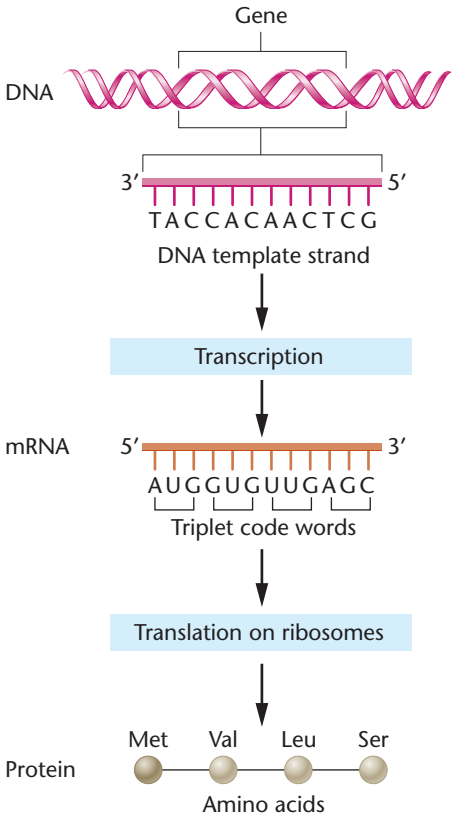


FIGURE 1.8 Gene expression consists of transcription of DNA into mRNA (top) and the translation (center) of mRNA (with the help of a ribosome) into a protein (bottom).

in DNA and specifies the insertion of a specific amino acid into a protein. Proteins (lower part of Figure 1.8) are polymers made up of amino acid monomers. There are 20 different amino acids commonly found in proteins.

Protein assembly is accomplished with the aid of adapter molecules called **transfer RNA (tRNA)**. Within the ribosome, tRNAs recognize the information encoded in the mRNA codons and carry the proper amino acids for construction of the protein during translation.

We now know that gene expression can be more complex than outlined here. Some of these complexities will be discussed later in the text (see Chapters 14 and 19).

Proteins and Biological Function

In most cases, proteins are the end products of gene expression. The diversity of proteins and the biological functions they perform—the diversity of life itself—arises from the fact that proteins are made from combinations of 20 different amino acids. Consider that a protein chain containing 100 amino acids can have at each position any one of 20 amino acids; the number of possible different 100-amino-acid proteins, each with a unique sequence, is therefore equal to

$$20^{100}$$

Obviously, proteins are molecules with the potential for enormous structural diversity and serve as the mainstay of biological systems.

Enzymes form the largest category of proteins. These molecules serve as biological catalysts, lowering the energy of activation in reactions and allowing cellular metabolism to proceed at body temperature.

Proteins other than enzymes are critical components of cells and organisms. These include hemoglobin, the oxygen-binding molecule in red blood cells; insulin, a pancreatic hormone; collagen, a connective tissue molecule; and actin and myosin, the contractile muscle proteins. A protein's shape and chemical behavior are determined by its linear sequence of amino acids, which in turn is dictated by the stored information in the DNA of a gene that is transferred to RNA, which then directs the protein's synthesis.

Linking Genotype to Phenotype: Sickle-Cell Anemia

Once a protein is made, its biochemical or structural properties play a role in producing a phenotype. When mutation alters a gene, it may modify or even eliminate the encoded protein's usual function and cause an altered phenotype. To trace this chain of events, we will examine sickle-cell anemia, a human genetic disorder.

Sickle-cell anemia is caused by a mutant form of hemoglobin, the protein that transports oxygen from the lungs to cells in the body. Hemoglobin is a composite molecule made up of two different proteins, α-globin and β-globin, each encoded by a different gene. In sickle-cell anemia,

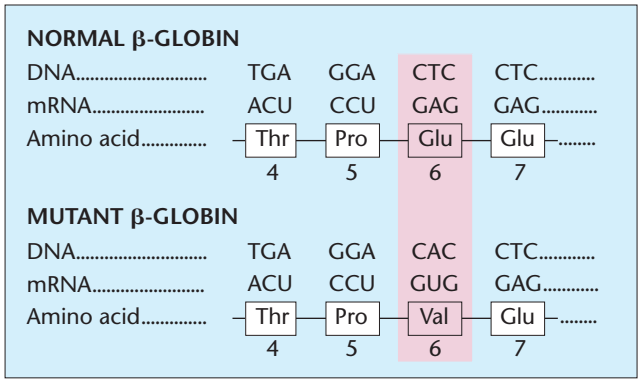


FIGURE 1.9 A single-nucleotide change in the DNA encoding β-globin (CTC → CAC) leads to an altered mRNA codon (GAG → GUG) and the insertion of a different amino acid (Glu → Val), producing the altered version of the β-globin protein that is responsible for sickle-cell anemia.

a mutation in the gene encoding β-globin causes an amino acid substitution in 1 of the 146 amino acids in the protein. **Figure 1.9** shows the DNA sequence, the corresponding mRNA codons, and the amino acids occupying positions 4–7 for the normal and mutant forms of β-globin. Notice that the mutation in sickle-cell anemia consists of a change in one DNA nucleotide, which leads to a change in codon 6 in mRNA from GAG to GUG, which in turn changes amino acid number 6 in β-globin from glutamic acid to valine. The other 145 amino acids in the protein are not changed by this mutation.

Individuals with two mutant copies of the β-globin gene have sickle-cell anemia. Their mutant β-globin proteins cause hemoglobin molecules in red blood cells to polymerize when the blood's oxygen concentration is low, forming long chains of hemoglobin that distort the shape of red blood cells (**Figure 1.10**). The deformed cells are fragile and break

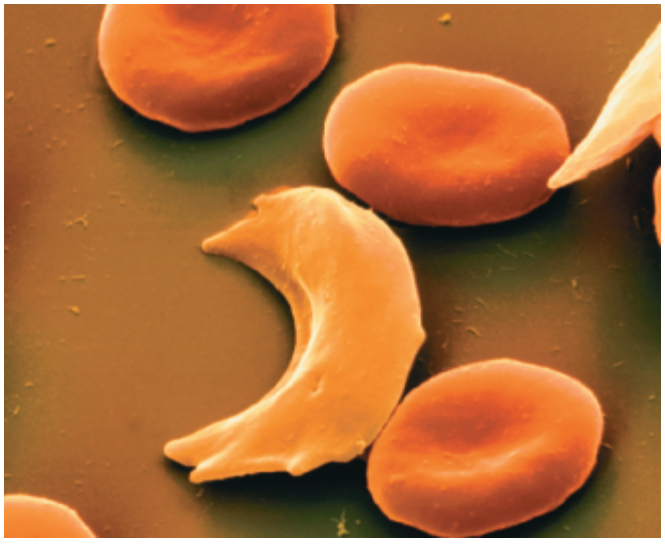


FIGURE 1.10 Normal red blood cells (round) and sickled red blood cells. The sickled cells block capillaries and small blood vessels.

easily, reducing the number of red blood cells in circulation (anemia is an insufficiency of red blood cells). Sickle-shaped blood cells block blood flow in capillaries and small blood vessels, causing severe pain and damage to the heart, brain, muscles, and kidneys. All the symptoms of this disorder are caused by a change in a single nucleotide in a gene that changes one amino acid out of 146 in the β-globin molecule, demonstrating the close relationship between genotype and phenotype.

1.4 Development of Recombinant DNA Technology Began the Era of DNA Cloning

The era of recombinant DNA began in the early 1970s, when researchers discovered that **restriction enzymes**, used by bacteria to cut and inactivate the DNA of invading viruses, could be used to cut any organism's DNA at specific nucleotide sequences, producing a reproducible set of fragments.

Soon after, researchers discovered ways to insert the DNA fragments produced by the action of restriction enzymes into carrier DNA molecules called **vectors** to form recombinant DNA molecules. When transferred into bacterial cells, thousands of copies, or **clones**, of the combined vector and DNA fragments are produced during bacterial reproduction. Large amounts of cloned DNA fragments can be isolated from these bacterial host cells. These DNA fragments can be used to isolate genes, to study their organization and expression, and to study their nucleotide sequence and evolution.

Collections of clones that represent an organism's **genome**, defined as the complete haploid DNA content of a specific organism, are called genomic libraries. Genomic libraries are now available for hundreds of species.

Recombinant DNA technology has not only accelerated the pace of research but also given rise to the biotechnology industry, which has grown to become a major contributor to the U.S. economy.

1.5 The Impact of Biotechnology Is Continually Expanding

The use of recombinant DNA technology and other molecular techniques to make products is called **biotechnology**. In the United States, biotechnology has quietly revolutionized many aspects of everyday life; products made by biotechnology are now found in the supermarket, in health care, in agriculture, and in the court system. A later chapter (see Chapter 22) contains a detailed discussion of biotechnology, but for now, let's look at some everyday examples of biotechnology's impact.

Plants, Animals, and the Food Supply

The use of recombinant DNA technology to genetically modify crop plants has revolutionized agriculture. Genes for traits including resistance to herbicides, insects, and genes for nutritional enhancement have been introduced into crop plants. The transfer of heritable traits across species using recombinant DNA technology creates **transgenic** organisms. Herbicide-resistant corn and soybeans were first planted in the mid-1990s, and transgenic strains now represent about 88 percent of the U.S. corn crop and 93 percent of the U.S. soybean crop. It is estimated that more than 70 percent of the processed food in the United States contains ingredients from transgenic crops.

We will discuss the most recent findings involving genetically modified organisms later in the text. (Special Topics Chapter 4—Genetically Modified Foods).

New methods of cloning livestock such as sheep and cattle have also changed the way we use these animals. In 1996, Dolly the sheep (**Figure 1.11**) was cloned by nuclear transfer, a method in which the nucleus of an adult cell is transferred into an egg that has had its nucleus removed. This method makes it possible to produce dozens or hundreds of genetically identical offspring with desirable traits and has many applications in agriculture, sports, and medicine.

Biotechnology has also changed the way human proteins for medical use are produced. Through use of gene transfer, transgenic animals now synthesize these therapeutic

FIGURE 1.11 Dolly, a Finn Dorset sheep cloned from the genetic material of an adult mammary cell, shown next to her first-born lamb, Bonnie.

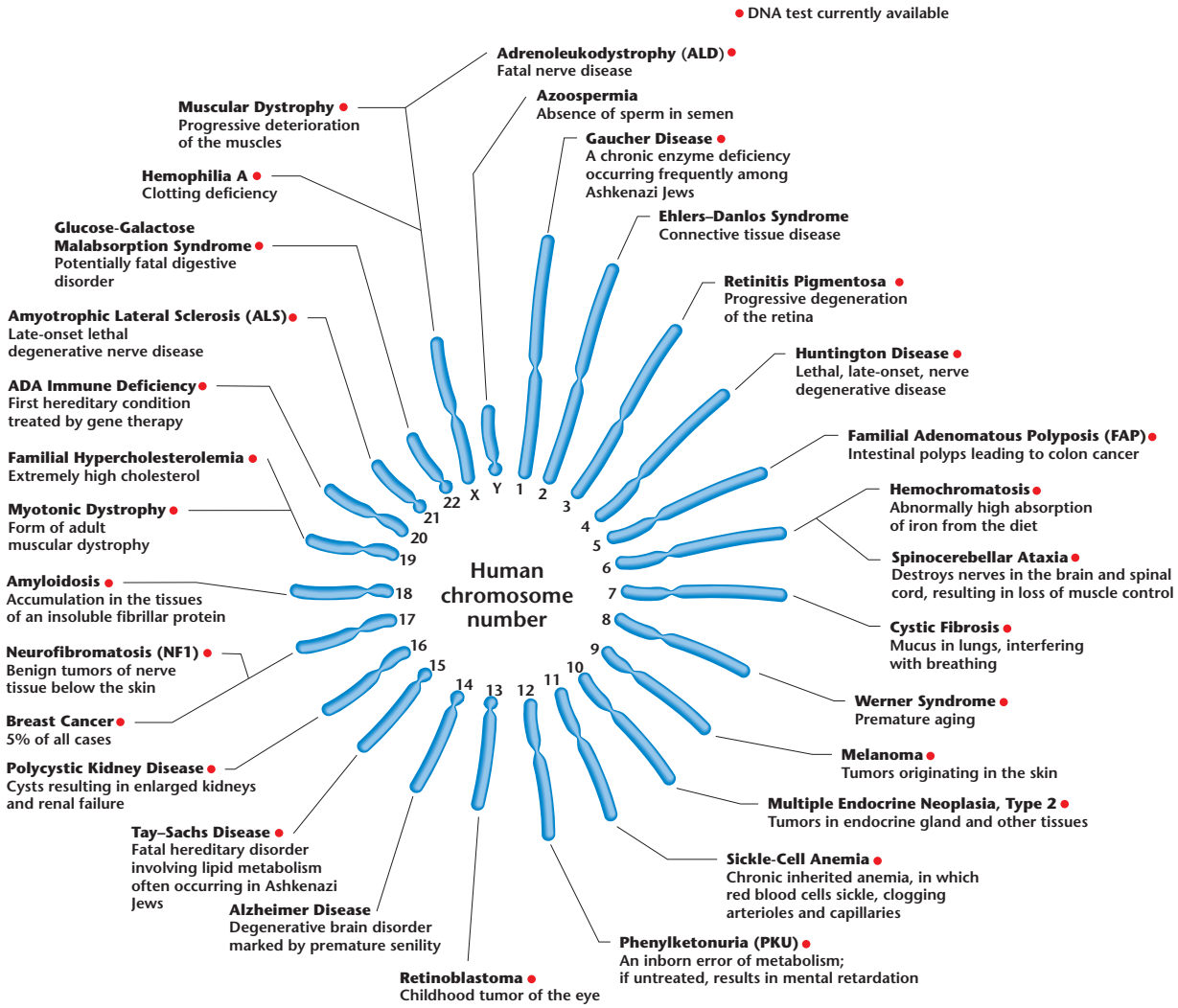


FIGURE 1.12 The human chromosome set, showing the location of some genes whose mutant forms cause hereditary diseases. Conditions that can be diagnosed using genetic testing are indicated by a red dot.

proteins. In 2009, an anticlotting protein derived from the milk of transgenic goats was approved by the U.S. Food and Drug Administration for use in the United States. Other human proteins from transgenic animals are now being used in clinical trials to treat several diseases. The biotechnology revolution will continue to expand as new methods are developed to make an increasing array of products.

Biotechnology in Genetics and Medicine

More than 10 million children or adults in the United States suffer from some form of genetic disorder, and every childbearing couple faces an approximately 3 percent risk of having a child with a genetic anomaly. The molecular basis for hundreds of genetic disorders is now known, and many of these genes have been mapped, isolated, and cloned (**Figure 1.12**). Biotechnology-derived genetic testing is now available to perform prenatal diagnosis of heritable disorders and to test parents for their status as "carriers" of more than 100 inherited disorders. Newer methods now under development offer the possibility of scanning an entire genome to establish an individual's risk of developing a genetic disorder or having an affected child. The use of genetic testing and related technologies raises ethical concerns that have yet to be resolved.

1.6 Genomics, Proteomics, and Bioinformatics Are New and Expanding Fields

The use of recombinant DNA technology to create genomic libraries prompted scientists to consider sequencing all the clones in a library to derive the nucleotide sequence of an organism's genome. This sequence information would be used to identify each gene in the genome and establish its function.

One such project, the Human Genome Project, began in 1990 as an international effort to sequence the human genome. By 2003, the publicly funded Human Genome Project and a private, industry-funded genome project completed sequencing of the gene-containing portion of the genome.

As more genome sequences were acquired, several new biological disciplines arose. One, called **genomics** (the study of genomes), studies the structure, function, and evolution of genes and genomes. A second field, **proteomics**, identifies the set of proteins present in a cell under a given set of conditions, and studies their functions and interactions. To store, retrieve, and analyze the massive amount of data generated by genomics and proteomics, a specialized subfield of information technology called **bioinformatics** was created to develop hardware and software for processing nucleotide and protein data.

Geneticists and other biologists now use information in databases containing nucleic acid sequences, protein sequences, and gene-interaction networks to answer experimental questions in a matter of minutes instead of months and years. A feature called "Exploring Genomics," located at the end of many of the chapters in this textbook, gives you the opportunity to explore these databases for yourself while completing an interactive genetics exercise.

Modern Approaches to Understanding Gene Function

This edition continues the feature "Modern Approaches to Understanding Gene Function" that appears in selected chapters. It is designed to introduce you to examples of the most current experimental approaches used by geneticists to study gene function. Its placement within these chapters links the techniques to the concepts that have just been presented.

Historically, an approach referred to as **classical** or **forward genetics** was essential for studying and understanding gene function. In this approach geneticists relied on the use of naturally occurring mutations or intentionally induced mutations (using chemicals, X-rays or UV light as examples) to cause altered phenotypes in model organisms, and then worked through the lab-intensive and time-consuming process of identifying the genes that caused these new phenotypes. Such characterization often led to the identification of the gene or genes of interest, and once the technology advanced, the gene sequence could be determined.

Classical genetics approaches are still used, but as whole genome sequencing has become routine, molecular approaches to understanding gene function have changed considerably in genetic research. These modern approaches are what we will highlight in this feature.

For the past two decades or so, geneticists have relied on the use of molecular techniques incorporating an approach referred to as **reverse genetics**. In reverse genetics, the DNA sequence for a particular gene of interest is known, but the role and function of the gene are typically not well understood. For example, molecular biology techniques such as **gene knockout** render targeted genes nonfunctional in a model organism or in cultured cells, allowing scientists to investigate the fundamental question of "what happens if this gene is disrupted?" After making a knockout organism, scientists look for both apparent phenotype changes, as well as those at the cellular and molecular level. The ultimate goal is to determine the function of the gene.

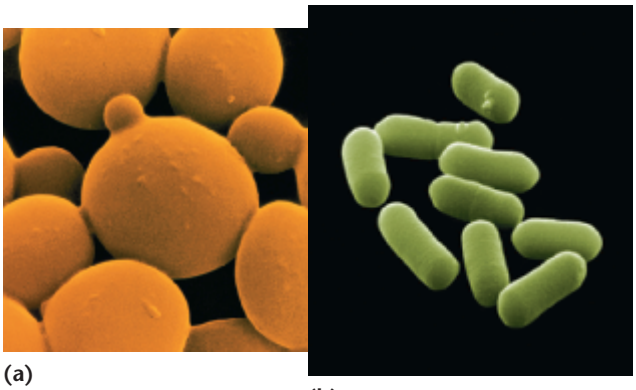
In "Modern Approaches to Understanding Gene Function" we will highlight experimental examples of how gene function has been revealed through modern applications of molecular techniques involving reverse genetics. You will learn about gene knockouts, transgenic animals, transposon-mediated mutagenesis, gene overexpression, and RNA interference-based methods for interrupting genes, among other approaches. Our hope is to bring you to the "cutting edge" of genetic studies.

1.7 Genetic Studies Rely on the Use of Model Organisms

After the rediscovery of Mendel's work in 1900, research using a wide range of organisms confirmed that the principles of inheritance he described were of universal significance among plants and animals. Geneticists gradually came to focus attention on a small number of organisms, including the fruit fly (*Drosophila melanogaster*) and the mouse (*Mus musculus*) (**Figure 1.13**). This trend developed for two main reasons: first, it was clear that genetic mechanisms were the same in most organisms, and second, these organisms had characteristics that made them especially suitable for genetic research. They were easy to grow, had relatively

(a) (b)

FIGURE 1.13 The first generation of model organisms in genetic analysis included (a) the mouse, *Mus musculus* and (b) the fruit fly, *Drosophila melanogaster*.



(a) (b)

FIGURE 1.14 Microbes that have become model organisms for genetic studies include (a) the yeast *Saccharomyces cerevisiae* and (b) the bacterium *Escherichia coli*.

short life cycles, produced many offspring, and their genetic analysis was fairly straightforward. Over time, researchers created a large catalog of mutant strains for these species, and the mutations were carefully studied, characterized, and mapped. Because of their well-characterized genetics, these species became **model organisms**, defined as organisms used for the study of basic biological processes. In later chapters, we will see how discoveries in model organisms are shedding light on many aspects of biology, including aging, cancer, the immune system, and behavior.

The Modern Set of Genetic Model Organisms

Gradually, geneticists added other species to their collection of model organisms: viruses (such as the T phages and lambda phage) and microorganisms (the bacterium *Escherichia coli* and the yeast *Saccharomyces cerevisiae*) (**Figure 1.14**).

More recently, additional species have been developed as model organisms, three of which are shown in the chapter opening photograph. Each species was chosen to allow study of some aspect of embryonic development. The nematode *Caenorhabditis elegans* was chosen as a model system to study the development and function of the nervous system because its nervous system contains only a few hundred cells and the developmental fate of these and all other cells in the body has been mapped out. *Arabidopsis thaliana*, a small plant with a short life cycle, has become a model organism for the study of many aspects of plant biology. The zebrafish, *Danio rerio*, is used to study vertebrate development: it is small, it reproduces rapidly, and its egg, embryo, and larvae are all transparent.

Model Organisms and Human Diseases

The development of recombinant DNA technology and the results of genome sequencing have confirmed that all life has a common origin. Because of this, genes with similar functions in different organisms tend to be similar or identical in structure and nucleotide sequence. Much of what scientists learn by studying the genetics of model organisms can therefore be applied to humans as the basis for understanding and treating human diseases. In addition, the ability to create transgenic organisms by transferring genes between species has enabled scientists to develop models of human diseases in organisms ranging from bacteria to fungi, plants, and animals (**Table 1.1**).

TABLE 1.1 Model Organisms Used to Study Some Human Diseases

Organism	Human Diseases
E. coli	Colon cancer and other cancers
S. cerevisiae	Cancer, Werner syndrome
D. melanogaster	Disorders of the nervous system, cancer
C. elegans	Diabetes
D. rerio	Cardiovascular disease
M. musculus	Lesch–Nyhan disease, cystic fibrosis, fragile-X syndrome, and many other diseases

The idea of studying a human disease such as colon cancer by using *E. coli* may strike you as strange, but the basic steps of DNA repair (a process that is defective in some forms of colon cancer) are the same in both organisms, and a gene involved in DNA repair (*mutL* in *E. coli* and *MLH1* in humans) is found in both organisms. More importantly, *E. coli* has the advantage of being easier to grow (the cells divide every 20 minutes), and researchers can easily create and study new mutations in the bacterial *mutL* gene in order to figure out how it works. This knowledge may eventually lead to the development of drugs and other therapies to treat colon cancer in humans.

The fruit fly, *Drosophila melanogaster*, is also being used to study a number of human diseases. Mutant genes have been identified in *D. melanogaster* that produce phenotypes with structural abnormalities of the nervous system and adult-onset degeneration of the nervous system. The information from genome-sequencing projects indicates that almost all these genes have human counterparts. For example, genes involved in a complex human disease of the retina called retinitis pigmentosa are identical to *Drosophila* genes involved in retinal degeneration. Study of these mutations in *Drosophila* is helping to dissect this complex disease and identify the function of the genes involved.

Another approach to studying diseases of the human nervous system is to transfer mutant human disease genes into *Drosophila* using recombinant DNA technology. The transgenic flies are then used for studying the mutant human genes themselves, other genes that affect the expression of the human disease genes, and the effects of therapeutic drugs on the action of those genes—all studies that are difficult or impossible to perform in humans. This gene transfer approach is being used to study almost

a dozen human neurodegenerative disorders, including Huntington disease, Machado–Joseph disease, myotonic dystrophy, and Alzheimer disease.

Throughout the following chapters, you will encounter these model organisms again and again. Remember each time you meet them that they not only have a rich history in basic genetics research but are also at the forefront in the study of human genetic disorders and infectious diseases. As discussed in the next section, however, we have yet to reach a consensus on how and when some of this technology will be accepted as safe and ethically acceptable.

1.8 We Live in the Age of Genetics

Mendel described his decade-long project on inheritance in pea plants in an 1865 paper presented at a meeting of the Natural History Society of Brünn in Moravia. Less than 100 years later, the 1962 Nobel Prize was awarded to James Watson, Francis Crick, and Maurice Wilkins for their work on the structure of DNA. This time span encompassed the years leading up to the acceptance of Mendel's work, the discovery that genes are on chromosomes, the experiments that proved DNA encodes genetic information, and the elucidation of the molecular basis for DNA replication. The rapid development of genetics from Mendel's monastery garden to the Human Genome Project and beyond is summarized in a timeline in **Figure 1.15**.

The Nobel Prize and Genetics

No other scientific discipline has experienced the explosion of information and the level of excitement generated by the discoveries in genetics. This impact is especially apparent in the list of Nobel Prizes related to genetics, beginning with those awarded in the early and mid-twentieth century and continuing into the present (see inside front cover). Nobel Prizes in Medicine or Physiology and Chemistry have been consistently awarded for work in genetics and related fields. One of the first such prizes awarded was given to Thomas H. Morgan in 1933 for his research on the chromosome theory of inheritance. That award was followed by many others, including prizes for the discovery of genetic recombination, the relationship between genes and proteins, the structure of DNA, and the genetic code. This trend has continued throughout the twentieth and twenty-first centuries. The advent of genomic studies and the applications of such findings will most certainly lead the way for future awards.

Genetics, Ethics, and Society

Just as there has never been a more exciting time to study genetics, the impact of this discipline on society has never been more profound. Genetics and its applications in biotechnology are developing much faster than the social conventions, public policies, and laws required to regulate their use. As a society, we are grappling with a host of sensitive genetics-related issues, including concerns about prenatal testing, genetic discrimination, ownership of genes, access to and safety of gene therapy, and genetic privacy. Two features appearing at the end of most chapters, "Case Study" and "Genetics, Ethics, and Society," consider ethical issues raised by the use of genetic technology. This emphasis on ethics reflects the growing concern and dilemmas that advances in genetics pose to our society and the future of our species.

By the time you finish this course, you will have seen more than enough evidence to convince yourself that the present is the Age of Genetics, and you will understand the need to think about and become a participant in the dialogue concerning genetic science and its use.

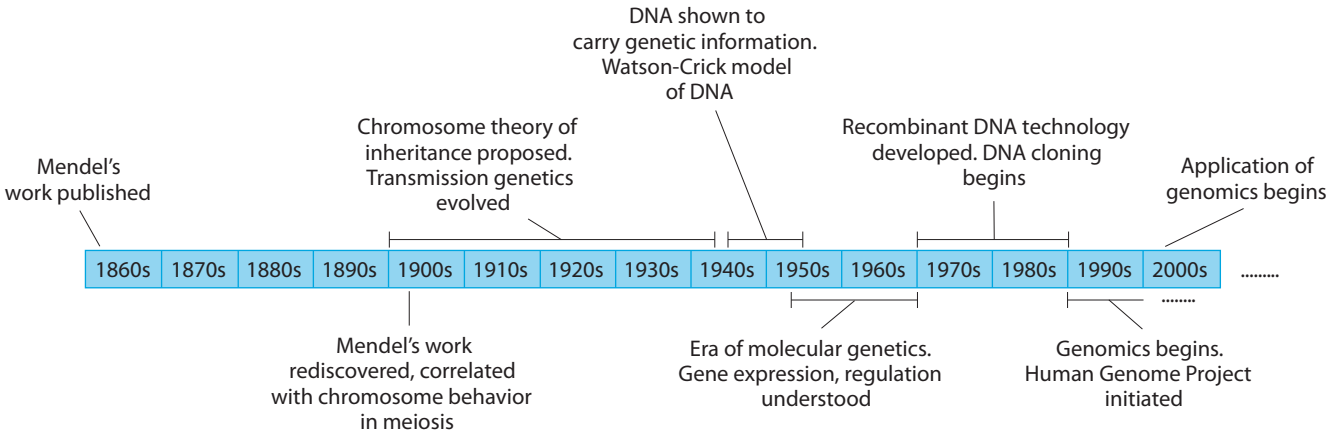


FIGURE 1.15 A timeline showing the development of genetics from Gregor Mendel's work on pea plants to the current era of genomics and its many applications in research, medicine, and society. Having a sense of the history of discovery in genetics should provide you with a useful framework as you proceed through this textbook.

Summary Points

Mastering Genetics For activities, animations, and review quizzes, go to the Study Area.

1. Mendel's work on pea plants established the principles of gene transmission from parents to offspring that form the foundation for the science of genetics.
2. Genes and chromosomes are the fundamental units in the chromosomal theory of inheritance. This theory explains that inherited traits are controlled by genes located on chromosomes and shows how the transmission of genetic information maintains genetic continuity from generation to generation.
3. Molecular genetics—based on the central dogma that DNA is a template for making RNA, which encodes the order of amino acids in proteins—explains the phenomena described by Mendelian genetics, referred to as transmission genetics.
4. Recombinant DNA technology, a far-reaching methodology used in molecular genetics, allows genes from one organism to be spliced into vectors and cloned, producing many copies of specific DNA sequences.
5. Biotechnology has revolutionized agriculture, the pharmaceutical industry, and medicine. It has made possible the mass production of medically important gene products. Genetic testing allows detection of individuals with genetic disorders and those at risk of having affected children, and gene therapy offers hope for the treatment of serious genetic disorders.
6. Genomics, proteomics, and bioinformatics are new fields derived from recombinant DNA technology. These fields combine genetics with information technology and allow scientists to explore genome sequences, the structure and function of genes, the protein set within cells, and the evolution of genomes. The Human Genome Project is one example of genomics.
7. The use of model organisms has advanced the understanding of genetic mechanisms and, coupled with recombinant DNA technology, has produced models of human genetic diseases.
8. The effects of genetic technology on society are profound, and the development of policy and legislation to deal with issues derived from the use of this technology is lagging behind the resulting innovations.

Problems and Discussion Questions

Mastering Genetics Visit for instructor-assigned tutorials and problems.

1. How does Mendel's work on the transmission of traits relate to our understanding of genetics today?

2. CONCEPT QUESTION Review the Chapter Concepts list on p. 37. Most of these are related to the discovery of DNA as the genetic material and the subsequent development of recombinant DNA technology. Write a brief essay that discusses the impact of recombinant DNA technology on genetics as we perceive the discipline today.

3. What is the chromosome theory of inheritance, and how is it related to Mendel's findings?

4. Define genotype and phenotype. Describe how they are related and how alleles fit into your definitions.

5. Given the state of knowledge at the time of the Avery, MacLeod, and McCarty experiment, why was it difficult for some scientists to accept that DNA is the carrier of genetic information?

6. What is a gene?

7. What is the structure of DNA? How does it differ from that of RNA?

8. Describe the central dogma of molecular genetics and how it serves as the basis of modern genetics.

9. Until the mid-1940s, many scientists considered proteins to be the likely candidates for the genetic material. Why?

10. Outline the roles played by restriction enzymes and vectors in cloning DNA.

11. What are some of the impacts of biotechnology on crop plants in the United States?

12. Summarize the arguments for and against patenting genetically modified organisms.

13. We all carry about 20,000 genes in our genome. So far, patents have been issued for more than 6000 of these genes. Do you think that companies or individuals should be able to patent human genes? Why or why not?

14. How has the use of model organisms advanced our knowledge of the genes that control human diseases?

15. If you knew that a devastating late-onset inherited disease runs in your family (in other words, a disease that does not appear until later in life) and you could be tested for it at the age of 20, would you want to know whether you are a carrier? Would your answer be likely to change when you reach age 40?

16. Why do you think discoveries in genetics have been recognized with so many Nobel Prizes?

17. The Age of Genetics was created by remarkable advances in the use of biotechnology to manipulate plant and animal genomes. Given that the world population reached 7.5 billion people in 2017 and is expected to reach 9.7 billion in 2050, some scientists have proposed that only the worldwide introduction of genetically modified (GM) foods will increase crop yields enough to meet future nutritional demands. Pest resistance, herbicide, cold, drought, and salinity tolerance, along with increased nutrition, are seen as positive attributes of GM foods. However, others caution that unintended harm to other organisms, reduced effectiveness to pesticides, gene transfer to nontarget species, allergenicity, and as yet unknown effects on human health are potential concerns regarding GM foods. If you were in a position to control the introduction of a GM primary food product (rice, for example), what criteria would you establish before allowing such introduction?

2

Mitosis and Meiosis

Chromosomes in the prometaphase stage of mitosis, derived from a cell in the flower of *Haemanthus*.

CHAPTER CONCEPTS

- Genetic continuity between generations of cells and between generations of sexually reproducing organisms is maintained through the processes of mitosis and meiosis, respectively.
- Diploid eukaryotic cells contain their genetic information in pairs of homologous chromosomes, with one member of each pair being derived from the maternal parent and one from the paternal parent.
- Mitosis provides a mechanism by which chromosomes, having been duplicated, are distributed into progeny cells during cell reproduction.
- Mitosis converts a diploid cell into two diploid daughter cells.
- The process of meiosis distributes one member of each homologous pair of chromosomes into each gamete or spore, thus reducing the diploid chromosome number to the haploid chromosome number.
- Meiosis generates genetic variability by distributing various combinations of maternal and paternal members of each homologous pair of chromosomes into gametes or spores.
- During the stages of mitosis and meiosis, the genetic material is condensed into discrete structures called chromosomes.

Every living thing contains a substance described as the genetic material. Except in certain viruses, this material is composed of the nucleic acid DNA. DNA has an underlying linear structure possessing segments called genes, the products of which direct the metabolic activities of cells. An organism's DNA, with its arrays of genes, is organized into structures called **chromosomes**, which serve as vehicles for transmitting genetic information. The manner in which chromosomes are transmitted from one generation of cells to the next and from organisms to their descendants must be exceedingly precise. In this chapter we consider exactly how genetic continuity is maintained between cells and organisms.

Two major processes are involved in the genetic continuity of nucleated cells: **mitosis** and **meiosis**. Although the mechanisms of the two processes are similar in many ways, the outcomes are quite different. Mitosis leads to the production of two cells, each with the same number of chromosomes as the parent cell. In contrast, meiosis reduces the genetic content and the number of chromosomes by precisely half. This reduction is essential if sexual reproduction is to occur without doubling the amount of genetic material in each new generation. Strictly speaking, mitosis is that portion of the cell cycle during which the hereditary components are equally partitioned into daughter cells. Meiosis is part of a special type of cell division that leads to the production of sex cells: **gametes** or **spores**. This process is an essential step in the transmission of genetic information from an organism to its offspring.

Normally, chromosomes are visible only during mitosis and meiosis. When cells are not undergoing division, the genetic material making up chromosomes unfolds and uncoils into a diffuse network within the nucleus, generally referred to as **chromatin**. Before describing mitosis and meiosis, we will briefly review the structure of cells, emphasizing components that are of particular significance to genetic function. We will also compare the structural differences between the prokaryotic (nonnucleated) cells of bacteria and the eukaryotic cells of higher organisms. We then devote the remainder of the chapter to the behavior of chromosomes during cell division.

2.1 Cell Structure Is Closely Tied to Genetic Function

Before 1940, our knowledge of cell structure was limited to what we could see with the light microscope. Around 1940, the transmission electron microscope was in its early stages of development, and by 1950, many details of cell ultrastructure had emerged. Under the electron microscope, cells were seen as highly varied, highly organized structures whose form and function are dependent on specific genetic expression by each cell type. A new world of whorled membranes, organelles, microtubules, granules, and filaments was revealed. These discoveries revolutionized thinking in the entire field of biology. Many cell components, such as the nucleolus, ribosome, and centriole, are involved directly or indirectly with genetic processes. Other components—the mitochondria and chloroplasts—contain their own unique genetic information. Here, we will focus primarily on those aspects of cell structure that relate to genetic study. The generalized animal cell shown in **Figure 2.1** illustrates most of the structures we will discuss.

All cells are surrounded by a *plasma membrane*, an outer covering that defines the cell boundary and delimits the cell from its immediate external environment. This membrane is not passive but instead actively controls the movement of materials into and out of the cell. In addition to this membrane, plant cells have an outer covering called the *cell wall* whose major component is a polysaccharide called *cellulose*.

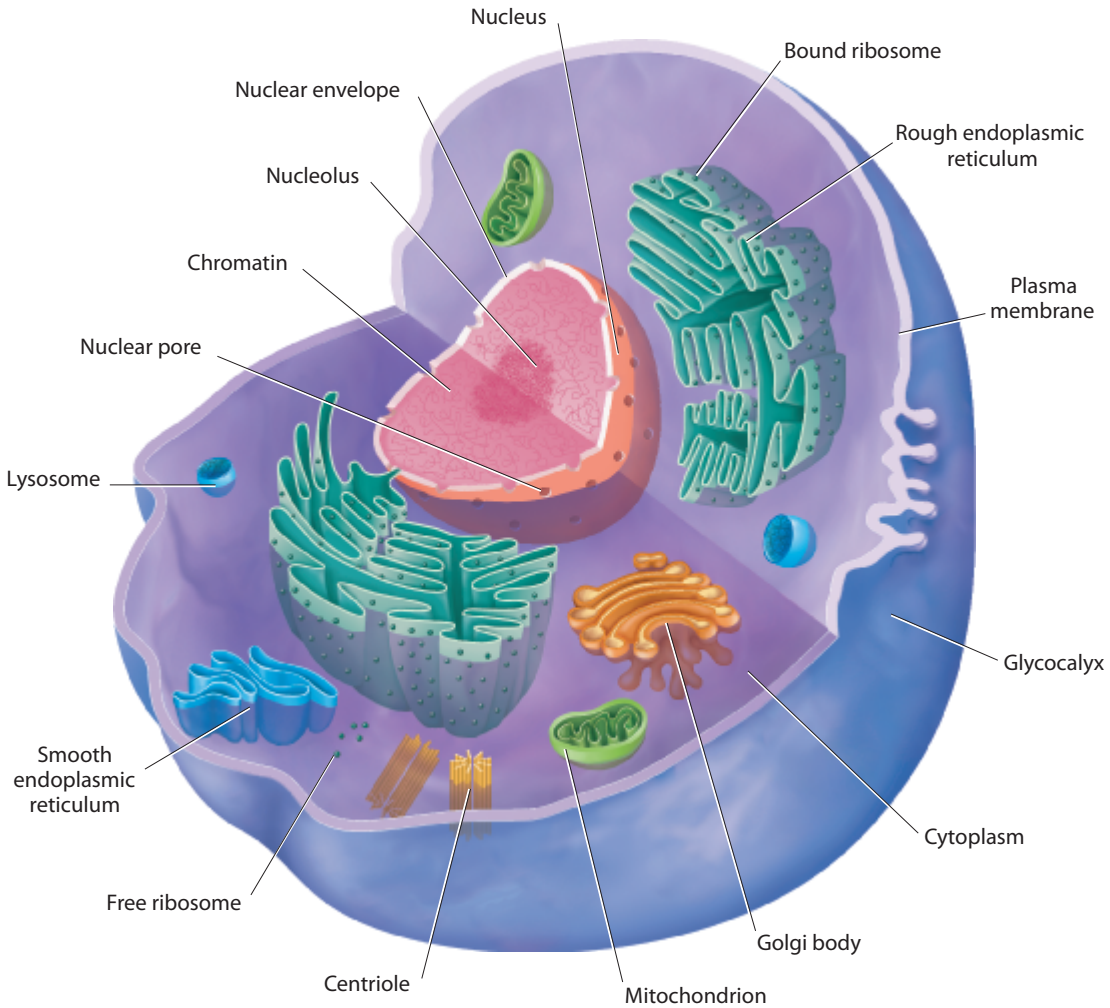


FIGURE 2.1 A generalized animal cell. The cellular components discussed in the text are emphasized here.

Many, if not most, animal cells have a covering over the plasma membrane, referred to as the **glycocalyx**, or *cell coat*. Consisting of glycoproteins and polysaccharides, this covering has a chemical composition that differs from comparable structures in either plants or bacteria. The glycocalyx, among other functions, provides biochemical identity at the surface of cells, and the components of the coat that establish cellular identity are under genetic control. For example, various cell-identity markers that you may have heard of—the AB, Rh, and MN antigens—are found on the surface of red blood cells, among other cell types. On the surface of other cells, histocompatibility antigens, which elicit an immune response during tissue and organ transplants, are present. Various **receptor molecules** are also found on the surfaces of cells. These molecules act as recognition sites that transfer specific chemical signals across the cell membrane into the cell.

Living organisms are categorized into two major groups depending on whether or not their cells contain a nucleus. The presence of a nucleus and other membranous organelles is the defining characteristic of **eukaryotic organisms**. The **nucleus** in eukaryotic cells is a membrane-bound structure that houses the genetic material, DNA, which is complexed with an array of acidic and basic proteins into thin fibers. During nondivisional phases of the cell cycle, the fibers are uncoiled and dispersed into chromatin (as mentioned above). During mitosis and meiosis, chromatin fibers coil and condense into chromosomes. Also present in the nucleus is the **nucleolus**, an amorphous component where ribosomal RNA (rRNA) is synthesized and where the initial stages of ribosomal assembly occur. The portions of DNA that encode rRNA are collectively referred to as the **nucleolus organizer region**, or the **NOR**.

Prokaryotic organisms, of which there are two major groups, lack a nuclear envelope and membranous organelles. For the purpose of our brief discussion here, we will consider the *eubacteria,* the other group being the more ancient bacteria referred to as *archaea.* In eubacteria, such as *Escherichia coli,* the genetic material is present as a long, circular DNA molecule that is compacted into an unenclosed region called the **nucleoid**. Part of the DNA may be attached to the cell membrane, but in general the nucleoid extends through a large part of the cell. Although the DNA is compacted, it does not undergo the extensive coiling characteristic of the stages of mitosis, during which the chromosomes of eukaryotes become visible. Nor is the DNA associated as extensively with proteins as is eukaryotic DNA. **Figure 2.2**, which shows two bacteria forming by cell division, illustrates the nucleoid regions containing the bacterial chromosomes. Prokaryotic cells do not have a distinct nucleolus but do contain genes that specify rRNA molecules.

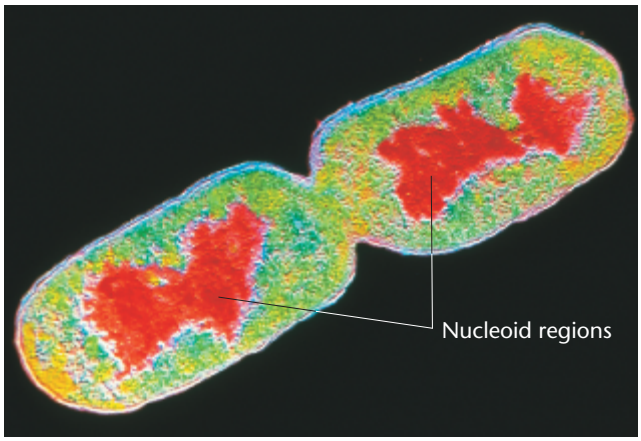


FIGURE 2.2 Color-enhanced electron micrograph of *E. coli* undergoing cell division. Particularly prominent are the two chromosomal areas (shown in red), called nucleoids, that have been partitioned into the daughter cells.

The remainder of the eukaryotic cell within the plasma membrane, excluding the nucleus, is referred to as **cytoplasm** and includes a variety of extranuclear cellular organelles. In the cytoplasm, a nonparticulate, colloidal material referred to as the *cytosol* surrounds and encompasses the cellular organelles. The cytoplasm also includes an extensive system of tubules and filaments, comprising the cytoskeleton, which provides a lattice of support structures within the cell. Consisting primarily of **microtubules**, which are made of the protein **tubulin**, and **microfilaments**, which derive from the protein *actin*, this structural framework maintains cell shape, facilitates cell mobility, and anchors the various organelles.

One organelle, the membranous **endoplasmic reticulum (ER)**, compartmentalizes the cytoplasm, greatly increasing the surface area available for biochemical synthesis. The ER appears smooth in places where it serves as the site for synthesizing fatty acids and phospholipids; in other places, it appears rough because it is studded with ribosomes. **Ribosomes** serve as sites where genetic information contained in messenger RNA (mRNA) is translated into proteins.

Three other cytoplasmic structures are very important in the eukaryotic cell's activities: mitochondria, chloroplasts, and centrioles. **Mitochondria** are found in most eukaryotes, including both animal and plant cells, and are the sites of the oxidative phases of cell respiration. These chemical reactions generate large amounts of the energy-rich molecule adenosine triphosphate (ATP). **Chloroplasts**, which are found in plants, algae, and some protozoans, are associated with photosynthesis, the major energy-trapping process on Earth. Both mitochondria and chloroplasts contain DNA in a form distinct from that found in the nucleus. They are able to duplicate themselves and transcribe and translate their own genetic information.

Animal cells and some plant cells also contain a pair of complex structures called **centrioles**. These cytoplasmic bodies, each located in a specialized region called the **centrosome**, are associated with the organization of spindle fibers that function in mitosis and meiosis. In some organisms, the centriole is derived from another structure, the basal body, which is associated with the formation of cilia and flagella (hair-like and whip-like structures for propelling cells or moving materials).

The organization of **spindle fibers** by the centrioles occurs during the early phases of mitosis and meiosis. These fibers play an important role in the movement of chromosomes as they separate during cell division. They are composed of arrays of microtubules consisting of polymers of the protein tubulin.

2.2 Chromosomes Exist in Homologous Pairs in Diploid Organisms

As we discuss the processes of mitosis and meiosis, it is important that you understand the concept of homologous chromosomes. Such an understanding will also be of critical importance in our future discussions of Mendelian genetics. Chromosomes are most easily visualized during mitosis. When they are examined carefully, distinctive lengths and shapes are apparent. Each chromosome contains a constricted region called the **centromere**, whose location establishes the general appearance of each chromosome. Figure 2.3 shows chromosomes with centromere placements at different distances along their length. Extending from either side of the centromere are the arms of the chromosome. Depending on the position of the centromere, different arm ratios are produced. As Figure 2.3 illustrates, chromosomes are classified as **metacentric**, **submetacentric**, **acrocentric**, or **telocentric** on the basis of the centromere location. The shorter arm, by convention, is shown above the centromere and is called the **p arm** (p, for "petite"). The longer arm is shown below the centromere and is called the **q arm** (q because it is the next letter in the alphabet).

In the study of mitosis, several other observations are of particular relevance. First, all somatic cells derived from members of the same species contain an identical number of chromosomes. In most cases, this represents what is referred to as the **diploid number (2*n*).** When the lengths and centromere placements of all such chromosomes are examined, a second general feature is apparent. With the exception of sex chromosomes, they exist in pairs with regard to these two properties, and the members of each pair are called **homologous chromosomes**. So, for each chromosome exhibiting a specific length and centromere placement, another exists with identical features.

There are exceptions to this rule. Many bacteria and viruses have but one chromosome, and organisms such as yeasts and molds, and certain plants such as bryophytes (mosses), spend the predominant phase of their life cycle in the haploid stage. That is, they contain only one member of each homologous pair of chromosomes during most of their lives.

Centromere location	Designation	Metaphase shape	Anaphase shape
Middle	Metacentric	Sister chromatids, Centromere	Migration
Between middle and end	Submetacentric	p arm, q arm	
Close to end	Acrocentric		
At end	Telocentric		

FIGURE 2.3 Centromere locations and the chromosome designations that are based on them. Note that the shape of the chromosome during anaphase is determined by the position of the centromere during metaphase.

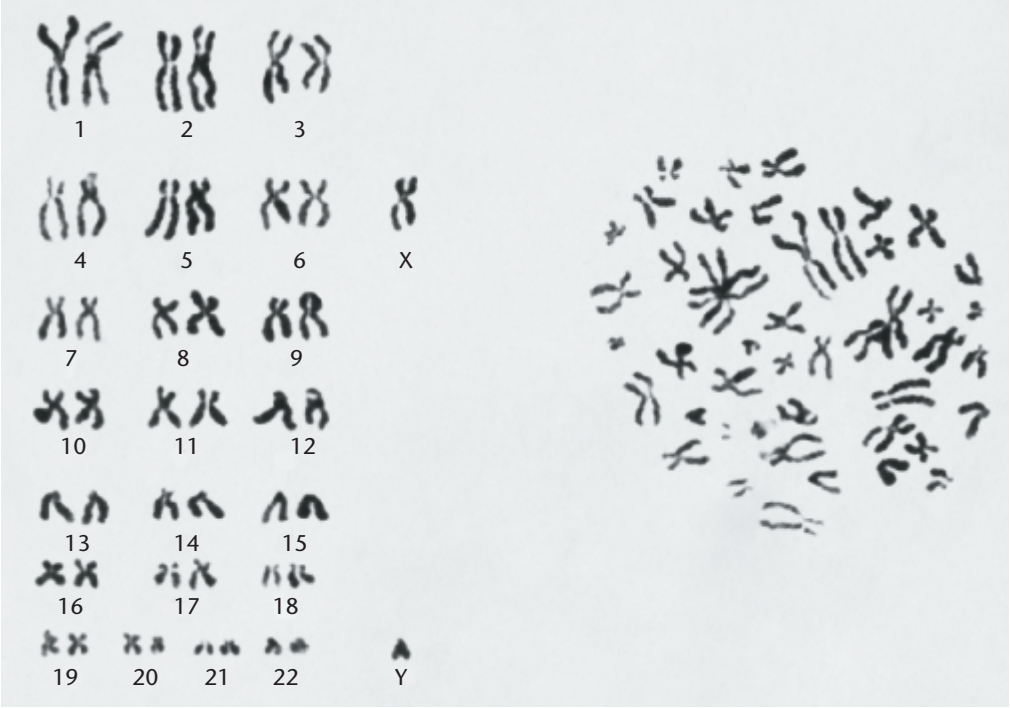


FIGURE 2.4 A metaphase preparation of chromosomes derived from a dividing cell of a human male (right), and the karyotype derived from the metaphase preparation (left). All but the X and Y chromosomes are present in homologous pairs. Each chromosome is clearly a double structure consisting of a pair of sister chromatids joined by a common centromere.

Figure 2.4 illustrates the physical appearance of different pairs of homologous chromosomes. There, the human mitotic chromosomes have been photographed, cut out of the print, and matched up, creating a display called a **karyotype**. As you can see, humans have a 2*n* number of 46 chromosomes, which on close examination exhibit a diversity of sizes and centromere placements. Note also that each of the 46 chromosomes in this karyotype is clearly a double structure consisting of two parallel *sister chromatids* connected by a common centromere. Had these chromosomes been allowed to continue dividing, the sister chromatids, which are replicas of one another, would have separated into the two new cells as division continued.

The **haploid number (*n*)** of chromosomes is equal to one-half the diploid number. Collectively, the genetic information contained in a haploid set of chromosomes constitutes the **genome** of the species. This, of course, includes copies of all genes as well as a large amount of noncoding DNA. The examples listed in **Table 2.1** demonstrate the wide range of *n* values found in plants and animals.

Homologous chromosomes have important genetic similarities. They contain identical gene sites along their lengths; each site is called a **locus** (pl. loci). Thus, they are identical in the traits that they influence and in their genetic potential. In sexually reproducing organisms, one member of each pair is derived from the maternal parent (through the ovum) and the other member is derived from the paternal parent (through the sperm). Therefore, each diploid organism contains two copies of each gene as a consequence of **biparental inheritance**, inheritance from two parents. As we shall see during our discussion of transmission genetics (Chapters 3 and 4), the members of each pair of genes, while influencing the same characteristic or trait, need not be identical. In a population of members of the same species, many different alternative forms of the same gene, called **alleles**, can exist.

TABLE 2.1 The Haploid Number of Chromosomes for a Variety of Organisms

Common Name	Scientific Name	Haploid Number
Black bread mold	*Aspergillus nidulans*	8
Broad bean	*Vicia faba*	6
Chimpanzee	*Pan troglodytes*	24
Corn	*Zea mays*	10
Cotton	*Gossypium hirsutum*	26
Dog	*Canis familiaris*	39
Fruit fly	*Drosophila melanogaster*	4
Garden pea	*Pisum sativum*	7
House mouse	*Mus musculus*	20
Human	*Homo sapiens*	23
Jimson weed	*Datura stramonium*	12
Pink bread mold	*Neurospora crassa*	7
Roundworm	*Caenorhabditis elegans*	6
Wheat	*Triticum aestivum*	21
Yeast	*Saccharomyces cerevisiae*	16
Zebrafish	*Danio rerio*	25

The concepts of haploid number, diploid number, and homologous chromosomes are important for understanding the process of meiosis. During the formation of gametes or spores, meiosis converts the diploid number of chromosomes to the haploid number. As a result, haploid gametes or spores contain precisely one member of each homologous pair of chromosomes—that is, one complete haploid set. Following fusion of two gametes at fertilization, the diploid number is reestablished; that is, the zygote contains two complete haploid sets of chromosomes. The constancy of genetic material is thus maintained from generation to generation.

There is one important exception to the concept of homologous pairs of chromosomes. In many species, one pair, consisting of the *sex-determining chromosomes,* is often not homologous in size, centromere placement, arm ratio, or genetic content. For example, in humans, while females carry two homologous X chromosomes, males carry one Y chromosome in addition to one X chromosome (Figure 2.4). These X and Y chromosomes are not strictly homologous. The Y is considerably smaller and lacks most of the gene loci contained on the X. Nevertheless, they contain homologous regions and behave as homologs in meiosis so that gametes produced by males receive either one X or one Y chromosome.

2.3 Mitosis Partitions Chromosomes into Dividing Cells

The process of mitosis is critical to all eukaryotic organisms. In some single-celled organisms, such as protozoans and some fungi and algae, mitosis (as a part of cell division) provides the basis for asexual reproduction. Multicellular diploid organisms begin life as single-celled fertilized eggs called **zygotes**. The mitotic activity of the zygote and the subsequent daughter cells is the foundation for the development and growth of the organism. In adult organisms, mitotic activity is the basis for wound healing and other forms of cell replacement in certain tissues. For example, the epidermal cells of the skin and the intestinal lining of humans are continuously sloughed off and replaced. Cell division also results in the continuous production of reticulocytes that eventually shed their nuclei and replenish the supply of red blood cells in vertebrates. In abnormal situations, somatic cells may lose control of cell division, and form a tumor.

The genetic material is partitioned into daughter cells during nuclear division, or **karyokinesis**. This process is quite complex and requires great precision. The chromosomes must first be exactly replicated and then accurately partitioned. The end result is the production of two daughter nuclei, each with a chromosome composition identical to that of the parent cell.

Karyokinesis is followed by cytoplasmic division, or **cytokinesis**. This less complex process requires a mechanism that partitions the volume into two parts and then encloses each new cell in a distinct plasma membrane. As the cytoplasm is reconstituted, organelles replicate themselves, arise from existing membrane structures, or are synthesized *de novo* (anew) in each cell.

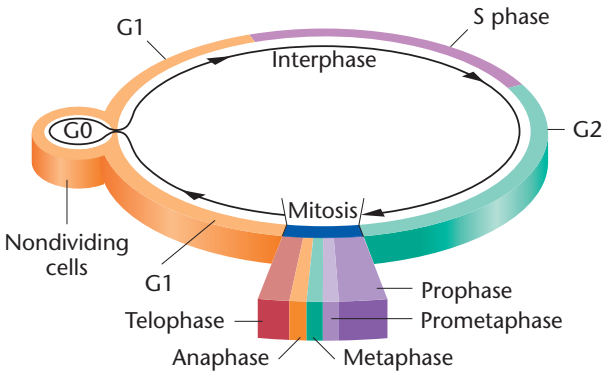


FIGURE 2.5 The stages comprising an arbitrary cell cycle. Following mitosis, cells enter the G1 stage of interphase, initiating a new cycle. Cells may become nondividing (G0) or continue through G1, where they become committed to begin DNA synthesis (S) and complete the cycle (G2 and mitosis). Following mitosis, two daughter cells are produced, and the cycle begins anew for both of them.

Following cell division, the initial size of each new daughter cell is approximately one-half the size of the parent cell. However, the nucleus of each new cell is not appreciably smaller than the nucleus of the original cell. Quantitative measurements of DNA confirm that there is an amount of genetic material in the daughter nuclei equivalent to that in the parent cell.

Interphase and the Cell Cycle

Many cells undergo a continuous alternation between division and nondivision. The events that occur from the completion of one division until the completion of the next division constitute the **cell cycle** (Figure 2.5). We will consider **interphase**, the initial stage of the cell cycle, as the interval between divisions. It was once thought that the biochemical activity during interphase was devoted solely to the cell's growth and its normal function. However, we now know that another biochemical step critical to the ensuing mitosis occurs during interphase: *the replication of the DNA of each chromosome.* This period, during which DNA is synthesized, occurs before the cell enters mitosis and is called the **S phase**. The initiation and completion of synthesis can be detected by monitoring the incorporation of radioactive precursors into DNA.

Investigations of this nature demonstrate two periods during interphase when no DNA synthesis occurs, one before and one after the S phase. These are designated **G1 (gap I)** and **G2 (gap II)**, respectively. During both of these intervals, as well as during S, intensive metabolic activity, cell growth, and cell differentiation are evident. By the end of G2, the volume of the cell has roughly doubled,

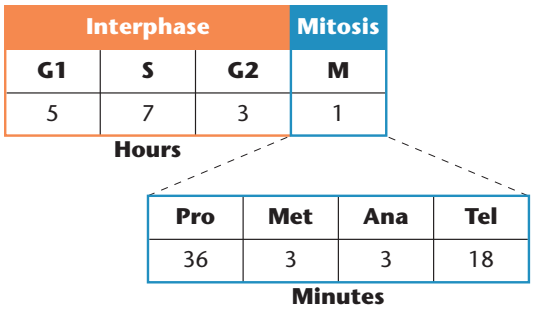


FIGURE 2.6 The time spent in each interval of one complete cell cycle of a human cell in culture. Times vary according to cell types and conditions.

DNA has been replicated, and mitosis (M) is initiated. Following mitosis, continuously dividing cells then repeat this cycle (G1, S, G2, M) over and over, as shown in Figure 2.5.

Much is known about the cell cycle based on *in vitro* (literally, "in glass") studies. When grown in culture, many cell types in different organisms traverse the complete cycle in about 16 hours. The actual process of mitosis occupies only a small part of the overall cycle, often less than an hour. The lengths of the S and G2 phases of interphase are fairly consistent in different cell types. Most variation is seen in the length of time spent in the G1 stage. **Figure 2.6** shows the relative length of these intervals as well as the length of the stages of mitosis in a human cell in culture.

G1 is of great interest in the study of cell proliferation and its control. At a point during G1, all cells follow one of two paths. They either withdraw from the cycle, become quiescent, and enter the **G0 stage** (see Figure 2.5), or they become committed to proceed through G1, initiating DNA synthesis, and completing the cycle. Cells that enter G0 remain viable and metabolically active but are not proliferative. Cancer cells apparently avoid entering G0 or pass through it very quickly. Other cells enter G0 and never reenter the cell cycle. Still other cells in G0 can be stimulated to return to G1 and thereby reenter the cell cycle.

Cytologically, interphase is characterized by the absence of visible chromosomes. Instead, the nucleus is filled with chromatin fibers that are formed as the chromosomes uncoil and disperse after the previous mitosis [**Figure 2.7(a)**]. Once G1, S, and G2 are completed, mitosis is initiated. Mitosis is a dynamic period of vigorous and continual activity. For discussion purposes, the entire process is subdivided into discrete stages, and specific events are assigned to each one. These stages, in order of occurrence, are prophase, prometaphase, metaphase, anaphase, and telophase. They are diagrammed with corresponding photomicrographs in Figure 2.7.

Prophase

Often, over half of mitosis is spent in **prophase** [**Figure 2.7(b)**], a stage characterized by several significant occurrences. One of the early events in prophase of all animal cells is the migration of two pairs of centrioles to opposite ends of the cell. These structures are found just outside the nuclear envelope in an area of differentiated cytoplasm called the centrosome (introduced in Section 2.1). It is believed that each pair of centrioles consists of one mature unit and a smaller, newly formed daughter centriole.

The centrioles migrate and establish poles at opposite ends of the cell. After migration, the centrosomes, in which the centrioles are localized, are responsible for organizing cytoplasmic microtubules into the spindle fibers that run between these poles, creating an axis along which chromosomal separation occurs. Interestingly, the cells of most plants (there are a few exceptions), fungi, and certain algae seem to lack centrioles. Spindle fibers are nevertheless apparent during mitosis.

As the centrioles migrate, the nuclear envelope begins to break down and gradually disappears. In a similar fashion, the nucleolus disintegrates within the nucleus. While these events are taking place, the diffuse chromatin fibers have begun to condense, until distinct thread-like structures, the chromosomes, become visible. It becomes apparent near the end of prophase that each chromosome is actually a double structure split longitudinally except at a single point of constriction, the centromere. The two parts of each chromosome are called **sister chromatids** because the DNA contained in each of them is genetically identical, having formed from a single replicative event. Sister chromatids are held together by a multi-subunit protein complex called **cohesin**. This molecular complex is originally formed between them during the S phase of the cell cycle when the DNA of each chromosome is replicated. Thus, even though we cannot see chromatids in interphase because the chromatin is uncoiled and dispersed in the nucleus, the chromosomes are already double structures, which becomes apparent in late prophase. In humans, with a diploid number of 46, a cytological preparation of late prophase reveals 46 chromosomes randomly distributed in the area formerly occupied by the nucleus.

Prometaphase and Metaphase

The distinguishing event of the two ensuing stages is the migration of every chromosome, led by its centromeric region, to the equatorial plane. The equatorial plane, also referred to as the *metaphase plate,* is the midline region of the cell, a plane that lies perpendicular to the axis established by the spindle fibers. In some descriptions, the term **prometaphase** refers to the period of chromosome movement [**Figure 2.7(c)**], and the term **metaphase** is applied strictly to the chromosome configuration following migration.

Migration is made possible by the binding of spindle fibers to the chromosome's **kinetochore**, an assembly of multilayered plates of proteins associated with the centromere. This structure forms on opposite sides of each paired centromere, in intimate association with the two sister

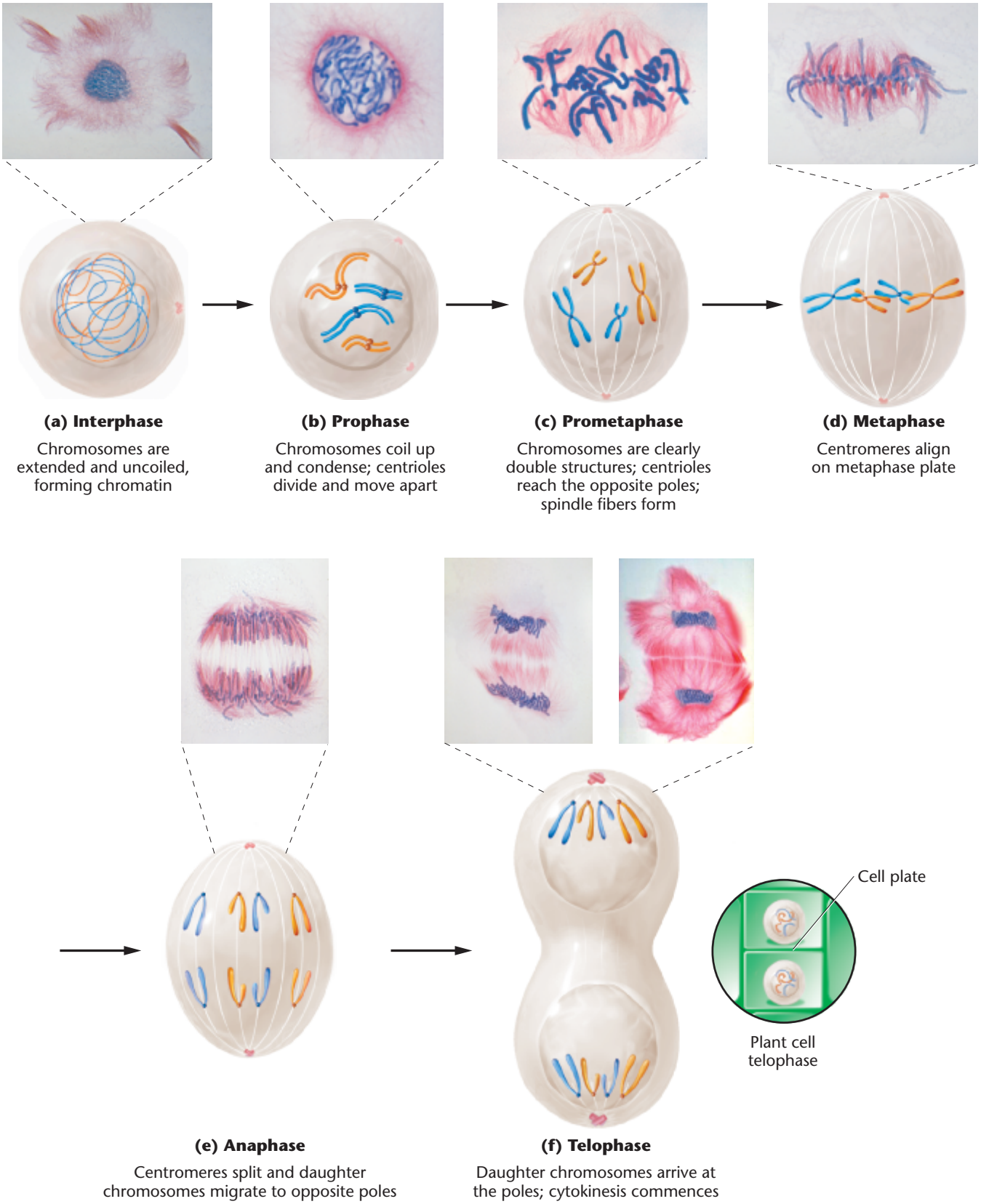


FIGURE 2.7 Drawings depicting mitosis in an animal cell with a diploid number of 4. The events occurring in each stage are described in the text. Of the two homologous pairs of chromosomes, one pair consists of longer, metacentric members and the other of shorter, submetacentric members. The maternal chromosome and the paternal chromosome of each pair are shown in different colors. To the right of (f), a drawing of late telophase in a plant cell shows the formation of the cell plate and lack of centrioles. The cells shown in the light micrographs came from the flower of *Haemanthus,* a plant that has a diploid number of 8.

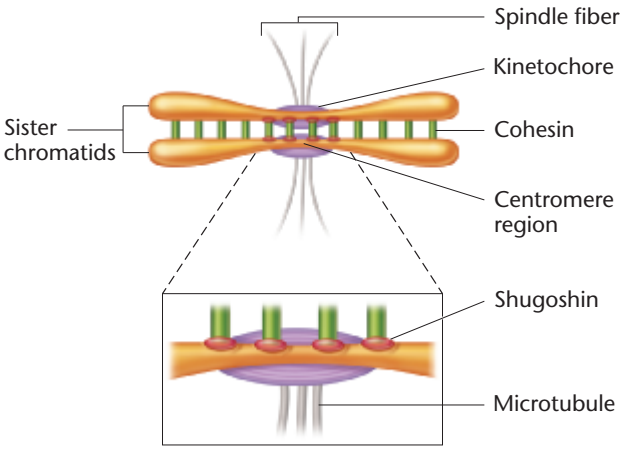


FIGURE 2.8 The depiction of the alignment, pairing, and disjunction of sister chromatids during mitosis, involving the molecular complexes cohesin and shugoshin and the enzyme separase.

chromatids. Once properly attached to the spindle fibers, cohesin is degraded by an enzyme, appropriately named *separase,* and the sister chromatid arms disjoin, except at the centromere region. A unique protein family called **shugoshin** (from the Japanese meaning "guardian spirit") protects cohesin from being degraded by separase at the centromeric regions. The involvement of the cohesin and shugoshin complexes with a pair of sister chromatids during mitosis is depicted in **Figure 2.8**.

We know a great deal about the molecular interactions involved in kinetechore assembly along the centromere. This is of great interest because of the consequences when mutations alter the proteins that make up the kinetechore complex. Altered kinetechore function potentially leads to errors during chromosome migration, altering the diploid content of daughter cells. A more detailed account will be presented later in the text, once we have provided more information about DNA and the proteins that make up chromatin (see Chapter 12).

We also know a great deal about spindle fibers and the mechanism responsible for their attachment to the kinetechore. Spindle fibers consist of microtubules, which themselves consist of molecular subunits of the protein tubulin. Microtubules seem to originate and "grow" out of the two centrosome regions at opposite poles of the cell. They are dynamic structures that lengthen and shorten as a result of the addition or loss of polarized tubulin subunits. The microtubules most directly responsible for chromosome migration make contact with, and adhere to, kinetochores as they grow from the centrosome region. They are referred to as *kinetochore microtubules* and have one end near the centrosome region (at one of the poles of the cell) and the other end anchored to the kinetochore. The number of microtubules that bind to the kinetochore varies greatly between organisms. Yeast (*Saccharomyces*) has only a single microtubule bound to each plate-like structure of the kinetochore. Mitotic cells of mammals, at the other extreme, reveal 30 to 40 microtubules bound to each portion of the kinetochore.

At the completion of metaphase, each centromere is aligned at the metaphase plate with the chromosome arms extending outward in a random array. This configuration is shown in **Figure 2.7(d)**.

Anaphase

Events critical to chromosome distribution during mitosis occur during **anaphase**, the shortest stage of mitosis. During this phase, sister chromatids of each chromosome, held together only at their centromere regions, *disjoin* (separate) from one another—an event described as **disjunction**—and are pulled to opposite ends of the cell. For complete disjunction to occur: (1) shugoshin must be degraded, reversing its protective role; (2) the cohesin complex holding the centromere region of each sister chromosome is then cleaved by separase; and (3) sister chromatids of each chromosome are pulled toward the opposite poles of the cell (Figure 2.8). As these events proceed, each migrating chromatid is now referred to as a *daughter chromosome.*

Movement of daughter chromosomes to the opposite poles of the cell is dependent on the kinetechore–spindle fiber attachment. Recent investigations reveal that chromosome migration results from the activity of a series of specific molecules called *motor proteins* found at several locations within the dividing cell. These proteins, described as **molecular motors**, use the energy generated by the hydrolysis of ATP. Their effect on the activity of microtubules serves ultimately to shorten the spindle fibers, drawing the chromosomes to opposite ends of the cell. The centromeres of each chromosome *appear* to lead the way during migration, with the chromosome arms trailing behind. Several models have been proposed to account for the shortening of spindle fibers. They share in common the selective removal of tubulin subunits at the ends of the spindle fibers. The removal process is accomplished by the molecular motor proteins described above.

The location of the centromere determines the shape of the chromosome during separation, as you saw in Figure 2.3. The steps that occur during anaphase are critical in providing each subsequent daughter cell with an identical set of chromosomes. In human cells, there would now be 46 chromosomes at each pole, one from each original sister pair. **Figure 2.7(e)** shows anaphase prior to its completion.

Telophase

Telophase is the final stage of mitosis and is depicted in **Figure 2.7(f)**. At its beginning, two complete sets of chromosomes are present, one set at each pole. The most significant event of this stage is cytokinesis, the division or partitioning of the cytoplasm. Cytokinesis is essential if two new cells are to be produced from one cell. The mechanism of cytokinesis differs greatly in plant and animal cells, but

the end result is the same: two new cells are produced. In plant cells, a *cell plate* is synthesized and laid down across the region of the metaphase plate. Animal cells, however, undergo a constriction of the cytoplasm, much as a loop of string might be tightened around the middle of a balloon.

It is not surprising that the process of cytokinesis varies in different organisms. Plant cells, which are more regularly shaped and structurally rigid, require a mechanism for depositing new cell wall material around the plasma membrane. The cell plate laid down during telophase becomes a structure called the *middle lamella*. Subsequently, the primary and secondary layers of the cell wall are deposited between the cell membrane and middle lamella in each of the resulting daughter cells. In animals, complete constriction of the cell membrane produces the *cell furrow* characteristic of newly divided cells.

Other events necessary for the transition from mitosis to interphase are initiated during late telophase. They generally constitute a reversal of events that occurred during prophase. In each new cell, the chromosomes begin to uncoil and become diffuse chromatin once again, while the nuclear envelope reforms around them, the spindle fibers disappear, and the nucleolus gradually reforms and becomes visible in the nucleus during early interphase. At the completion of telophase, the cell enters interphase.

Cell-Cycle Regulation and Checkpoints

The cell cycle, culminating in mitosis, is fundamentally the same in all eukaryotic organisms. This similarity in many diverse organisms suggests that the cell cycle is governed by a genetically regulated program that has been conserved throughout evolution. Because disruption of this regulation may underlie the uncontrolled cell division characterizing malignancy, interest in how genes regulate the cell cycle is particularly strong.

A mammoth research effort over the past 20 years has paid high dividends, and we now have knowledge of many genes involved in the control of the cell cycle. This work was recognized by the awarding of the 2001 Nobel Prize in Medicine or Physiology to Lee Hartwell, Paul Nurse, and Tim Hunt. As with other studies of genetic control over essential biological processes, investigation has focused on the discovery of mutations that interrupt the cell cycle and on the effects of those mutations. As we shall return to this subject in much greater detail later in the text during our consideration of the molecular basis of cancer (see Chapter 24), what follows is a very brief overview.

Many mutations are now known that exert an effect at one or another stage of the cell cycle. First discovered in yeast, but now evident in all organisms, including humans, such mutations were originally designated as *cell division cycle (cdc) mutations.* The normal products of many of the mutated genes are enzymes called **kinases** that can add phosphates to other proteins. They serve as "master control" molecules functioning in conjunction with proteins called **cyclins**. Cyclins bind to these kinases (creating *cyclin-dependent kinases*), activating them at appropriate times during the cell cycle. Activated kinases then phosphorylate other target proteins that regulate the progress of the cell cycle. The study of *cdc* mutations has established that the cell cycle contains at least three **cell-cycle checkpoints,** where the processes culminating in normal mitosis are monitored, or "checked," by these master control molecules before the next stage of the cycle is allowed to commence.

The importance of cell-cycle control and these checkpoints can be demonstrated by considering what happens when this regulatory system is impaired. Let's assume, for example, that the DNA of a cell has incurred damage leading to one or more mutations impairing cell-cycle control. If allowed to proceed through the cell cycle, this genetically altered cell would divide uncontrollably—a key step in the development of a cancer cell. If, instead, the cell cycle is arrested at one of the checkpoints, the cell can repair the DNA damage or permanently stop the cell from dividing, thereby preventing its potential malignancy. The specific checkpoints will be discussed in more detail later in the text (Chapter 24, Cancer Genetics).

NOW SOLVE THIS

2.1 With the initial appearance of the feature we call "Now Solve This," a short introduction is in order. The feature occurs several times in this and all ensuing chapters, each time providing a problem related to the discussion just presented. A "Hint" is then offered that may help you solve the problem. Here is the first problem:

(a) If an organism has a diploid number of 16, how many chromatids are visible at the end of mitotic prophase?

(b) How many chromosomes are moving to each pole during anaphase of mitosis?

■ **HINT:** *This problem involves an understanding of what happens to each pair of homologous chromosomes during mitosis, asking you to apply your understanding of chromosome behavior to an organism with a diploid number of 16. The key to its solution is your awareness that throughout mitosis, the members of each homologous pair do not pair up, but instead behave independently.*

2.4 Meiosis Creates Haploid Gametes and Spores and Enhances Genetic Variation in Species

Whereas in diploid organisms, mitosis produces two daughter cells with full diploid complements, **meiosis** produces gametes or spores that are characterized by only one haploid set of chromosomes. During sexual reproduction, haploid gametes then combine at fertilization to reconstitute the

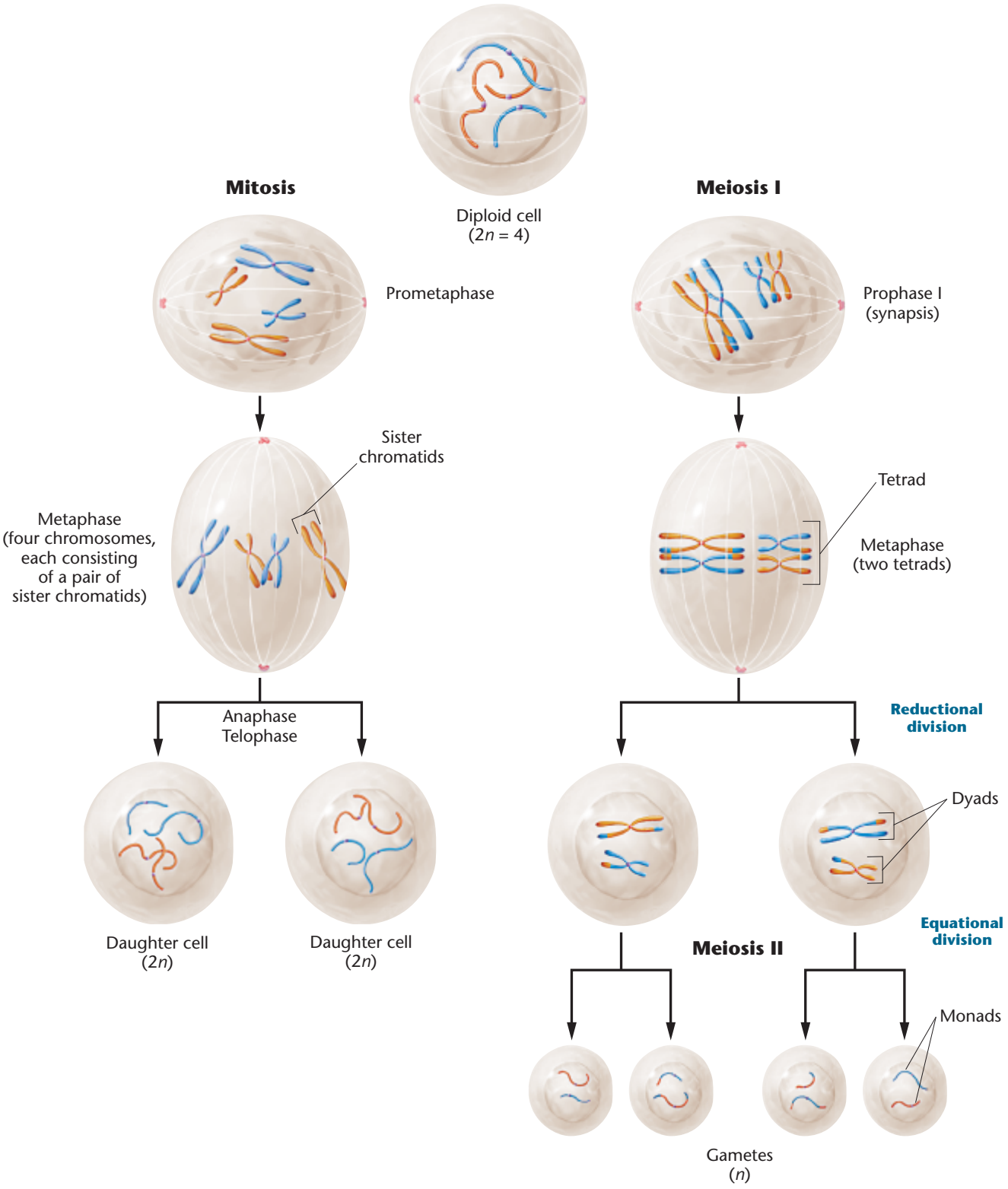


FIGURE 2.9 Overview of the major events and outcomes of mitosis and meiosis. As in Figure 2.7, two pairs of homologous chromosomes are followed.

diploid complement found in parental cells. **Figure 2.9** compares the two processes by following two pairs of homologous chromosomes. Meiosis must be highly specific since, by definition, haploid gametes or spores must contain precisely one member of each homologous pair of chromosomes. When successfully completed, meiosis provides the basis for maintaining genetic continuity from generation to generation.

Another major accomplishment of meiosis is to ensure that during sexual reproduction an enormous amount of genetic variation is produced among members of a species.

Such variation occurs in two forms. First, meiosis produces gametes with many unique combinations of maternally and paternally derived chromosomes among the haploid complement, thus ensuring that following fertilization, a large number of unique chromosome combinations are possible. As we will see (Chapter 3), this process is the underlying basis of Mendel's principles of segregation and independent assortment. The second source of variation is created by the meiotic event referred to as **crossing over**, which results in genetic exchange between members of each homologous pair of chromosomes prior to one or the other finding its way into a haploid gamete or spore. This creates intact chromosomes that are mosaics of the maternal and paternal homologs from which they arise, further enhancing genetic variation. Sexual reproduction therefore significantly reshuffles the genetic material, producing highly diverse offspring.

NOW SOLVE THIS

2.2 An organism has a diploid number of 16 in a primary oocyte. (a) How many tetrads are present in the first meiotic prophase? (b) How many dyads are present in the second meiotic prophase? (c) How many monads migrate to each pole during the second meiotic anaphase?

■ **HINT:** *This problem involves an understanding of what happens to the maternal and paternal members of each pair of homologous chromosomes during meiosis, asking you to apply your understanding of chromosome behavior in an organism with a diploid number of 16. The key to its solution is your awareness that maternal and paternal homologs synapse during meiosis. Once each chromatid has duplicated, creating a tetrad in the early phases of meiosis, each original pair behaves as a unit and leads to two dyads during anaphase I.*

For more practice, see Problems 25–30.

Meiosis: Prophase I

As in mitosis, the process in meiosis begins with a diploid cell duplicating its genetic material in the interphase stage preceding chromosome division. To achieve haploidy, two divisions are thus required. The meiotic achievements, as described above, are largely dependent on the behavior of chromosomes during the initial stage of the first division, called *prophase I.* Recall that in mitosis the paternally and maternally derived members of each homologous pair of chromosomes behave autonomously during division. Each chromosome is duplicated, creating genetically identical sister chromatids, and subsequently, one chromatid of each pair is distributed to each new cell. The major difference in meiosis is that once the chromatin characterizing interphase has condensed into visible structures, the homologous chromosomes are not autonomous but are instead seen to be paired up, having undergone the process called **synapsis**. **Figure 2.10** illustrates this process as well as the ensuing events of prophase I. Each synapsed pair of homologs is initially called a **bivalent,** and the number of bivalents is equal to the haploid number. In Figure 2.10, we have depicted two homologous pairs of chromosomes and thus two bivalents. As the homologs condense and shorten, each bivalent gives rise to a unit called a **tetrad,** consisting of two pairs of sister chromatids, each of which is joined at a common centromere. Remember that one pair of sister chromatids is maternally derived and the other pair is paternally derived. The presence of tetrads is visible evidence that *both* homologs have, in fact, duplicated. As prophase I progresses, each pair of sister chromatids within a tetrad is seen to pull apart. However, one or more areas remain in contact where chromatids are intertwined. Each such area, called a **chiasma** (pl., chiasmata), is thought to represent a point where **nonsister chromatids** (one paternal and one maternal chromatid) have undergone genetic exchange through the process of crossing over. Since crossing over is thought to occur one or more times in each tetrad, mosaic chromosomes are routinely created during every meiotic event. During the final period of prophase I, the nucleolus and nuclear envelope break down, and the two centromeres of each tetrad attach to the recently formed spindle fibers.

Metaphase, Anaphase, and Telophase I

The remainder of the meiotic process is depicted in **Figure 2.11**. After meiotic prophase I, stages similar to those of mitosis occur. In the first division, *metaphase I,* the chromosomes have maximally shortened and thickened.

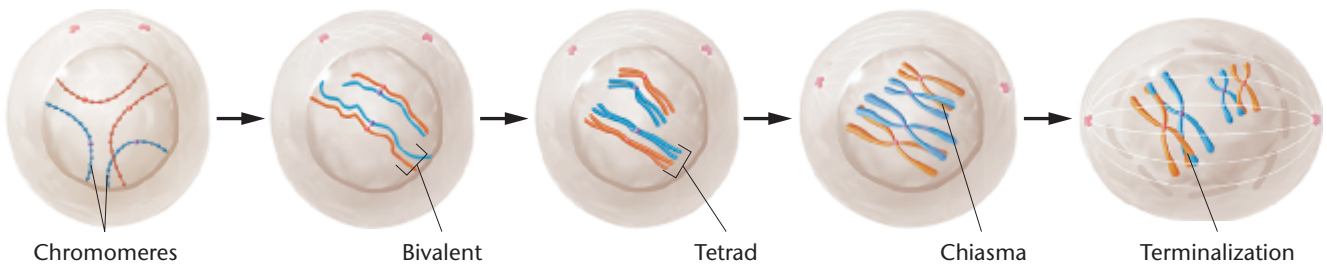


FIGURE 2.10 The changes in chromosome structure during prophase I. In the first two frames, illustrating chromomeres and bivalents, each chromatid is actually a double structure, consisting of sister chromatids, which first becomes apparent in the ensuing tetrad stage.

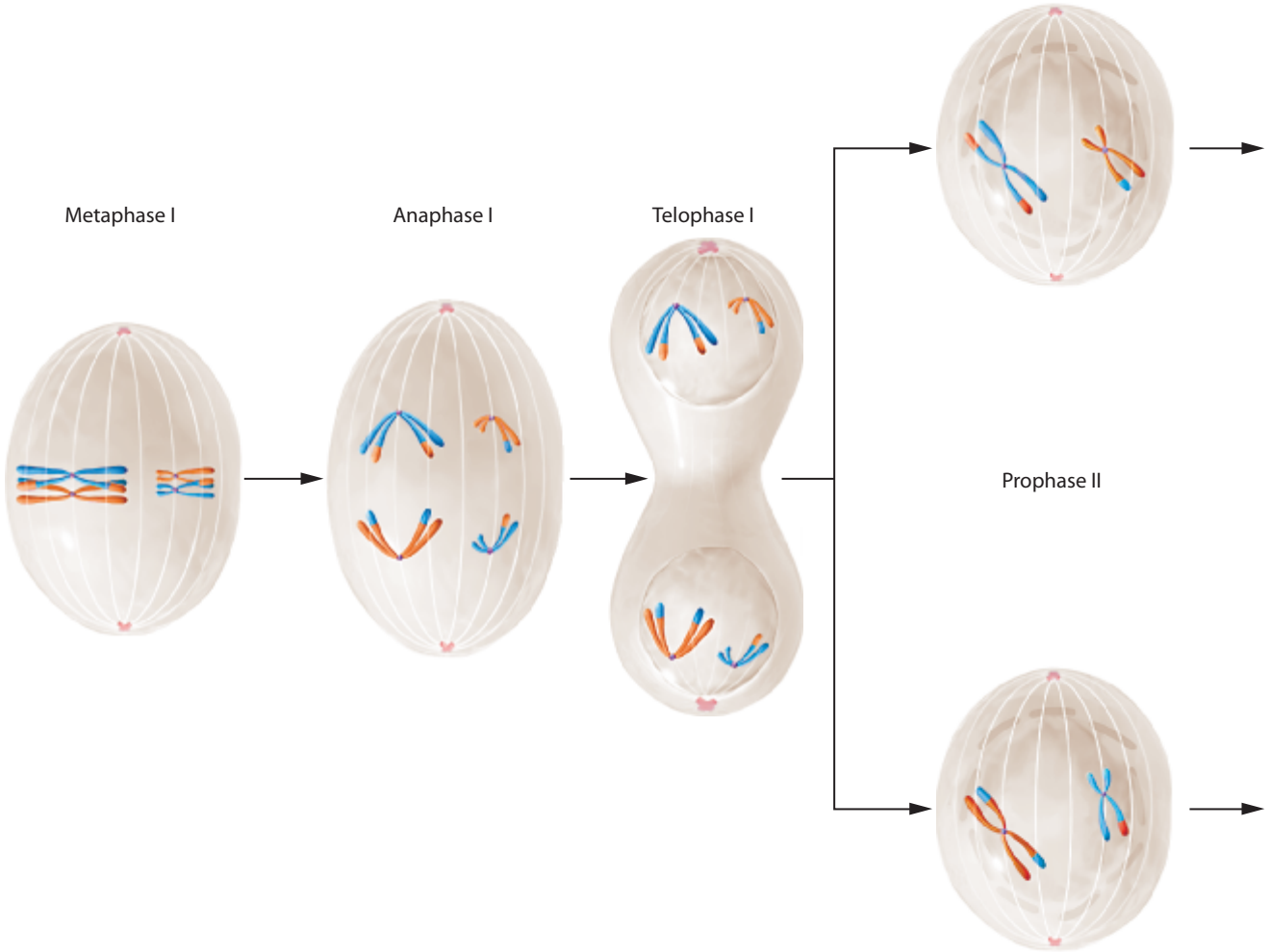


FIGURE 2.11 The major events in meiosis in an animal cell with a diploid number of 4, beginning with metaphase I. Note that the combination of chromosomes in the cells produced following telophase II is dependent on the random alignment of each tetrad and dyad on the equatorial plate during metaphase I and metaphase II. Several other combinations, which are not shown, can also be formed. The events depicted here are described in the text.

The terminal chiasmata of each tetrad are visible and appear to be the major factor holding the nonsister chromatids together. Each tetrad interacts with spindle fibers, facilitating its movement to the metaphase plate. The alignment of each tetrad prior to the first anaphase is random: Half of the tetrad (one of the dyads) will subsequently be pulled by spindle fibers to one or the other pole, and the other half will be pulled to the opposite pole.

During the stages of meiosis I, a single centromeric region holds each pair of sister chromatids together. It appears as a single unit, and a kinetechore forms around each one. As in our discussion of mitosis (see Figure 2.8), cohesin plays the major role in keeping sister chromatids together. At *anaphase I,* cohesin is degraded between sister chromatids, except at the centromere region, which, as in mitosis, is protected by a shugoshin complex. Then, one-half of each tetrad (a **dyad**) is pulled toward each pole of the dividing cell. Because this process effectively reduces the number of centromeres by half, it is referred to as a *reductional division*. This separation process is the physical basis of disjunction, the separation of homologous chromosomes from one another. Occasionally, errors in meiosis occur and separation is not achieved. The term **nondisjunction** describes such an error. At the completion of the normal anaphase I, a series of dyads equal to the haploid number is present at each pole.

If crossing over had not occurred in the first meiotic prophase, each dyad at each pole would consist solely of either paternal or maternal chromatids. However, the exchanges produced by crossing over create mosaic chromatids of paternal and maternal origin.

In many organisms, *telophase I* reveals a nuclear membrane forming around the dyads. In this case, the nucleus next

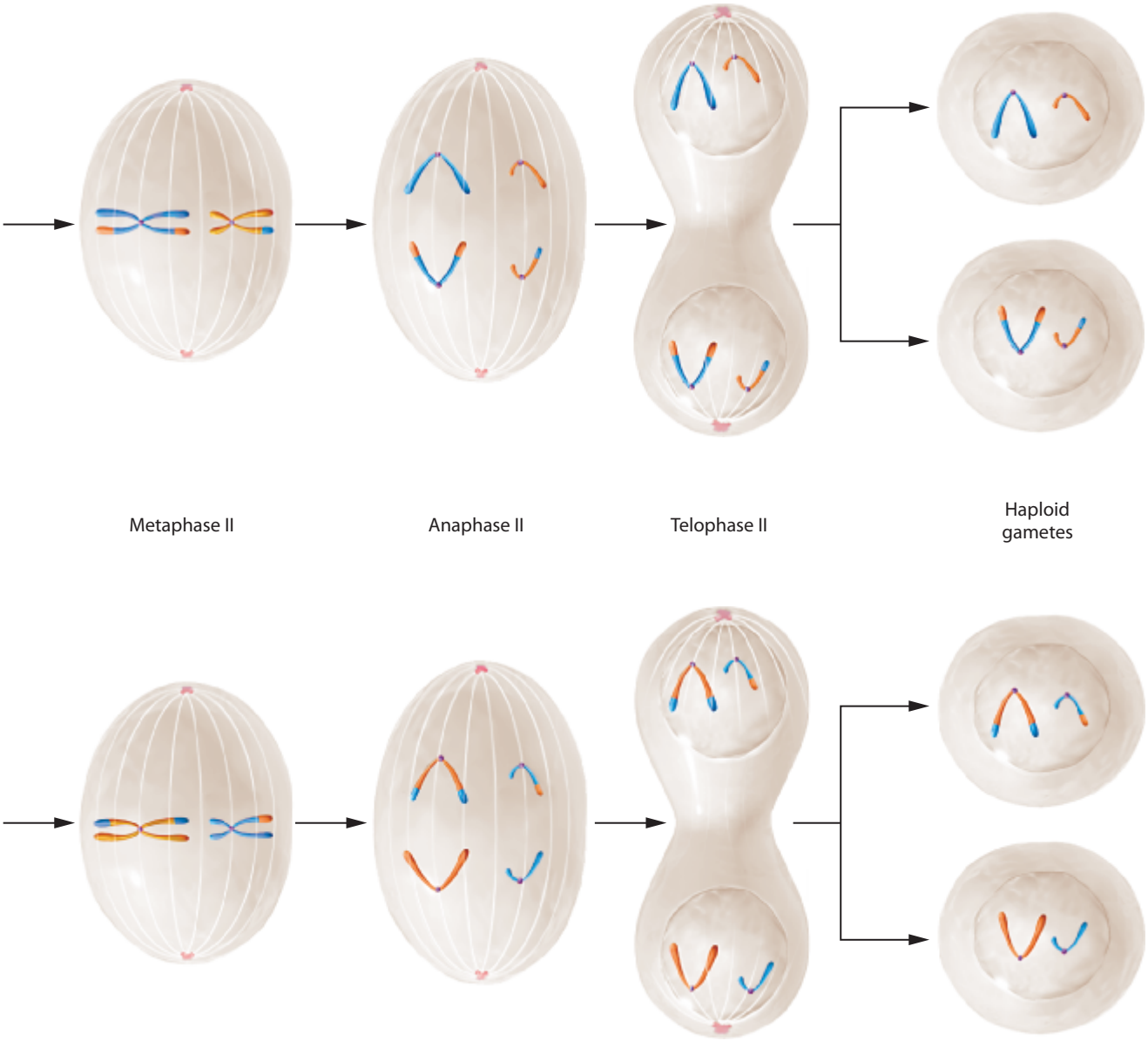


FIGURE 2.11 *(Continued)*

enters into a short interphase period. If interphase occurs, the chromosomes do not replicate because they already consist of two chromatids. In other organisms, the cells go directly from anaphase I to meiosis II. In general, meiotic telophase is much shorter than the corresponding stage in mitosis.

The Second Meiotic Division

A second division, referred to as *meiosis II,* is essential if each gamete or spore is to receive only one chromatid from each original tetrad. The stages characterizing meiosis II are shown on the right side of Figure 2.11. During *prophase II,* each dyad is composed of one pair of sister chromatids attached by the common centromeric region. During *metaphase II,* the centromeres are positioned on the equatorial plate. When the shugoshin complex is degraded, the centromeres separate, *anaphase II* is initiated, and the sister chromatids of each dyad are pulled to opposite poles. Because the number of dyads is equal to the haploid number, *telophase II* reveals one member of each pair of homologous chromosomes present at each pole. Each chromosome is now a monad. Because the number of centromeres is not reduced in number in the two resulting cells, the process is referred to as an *equational division*.

Following cytokinesis in telophase II, four haploid gametes may result from a single meiotic event. At the conclusion of meiosis II, not only has the haploid state been achieved, but if crossing over has occurred, each monad contains a combination of maternal and paternal genetic information. As a result, the offspring produced by any gamete will receive a mixture of genetic information originally present in his or her grandparents. Meiosis thus significantly increases the level of genetic variation in each ensuing generation.

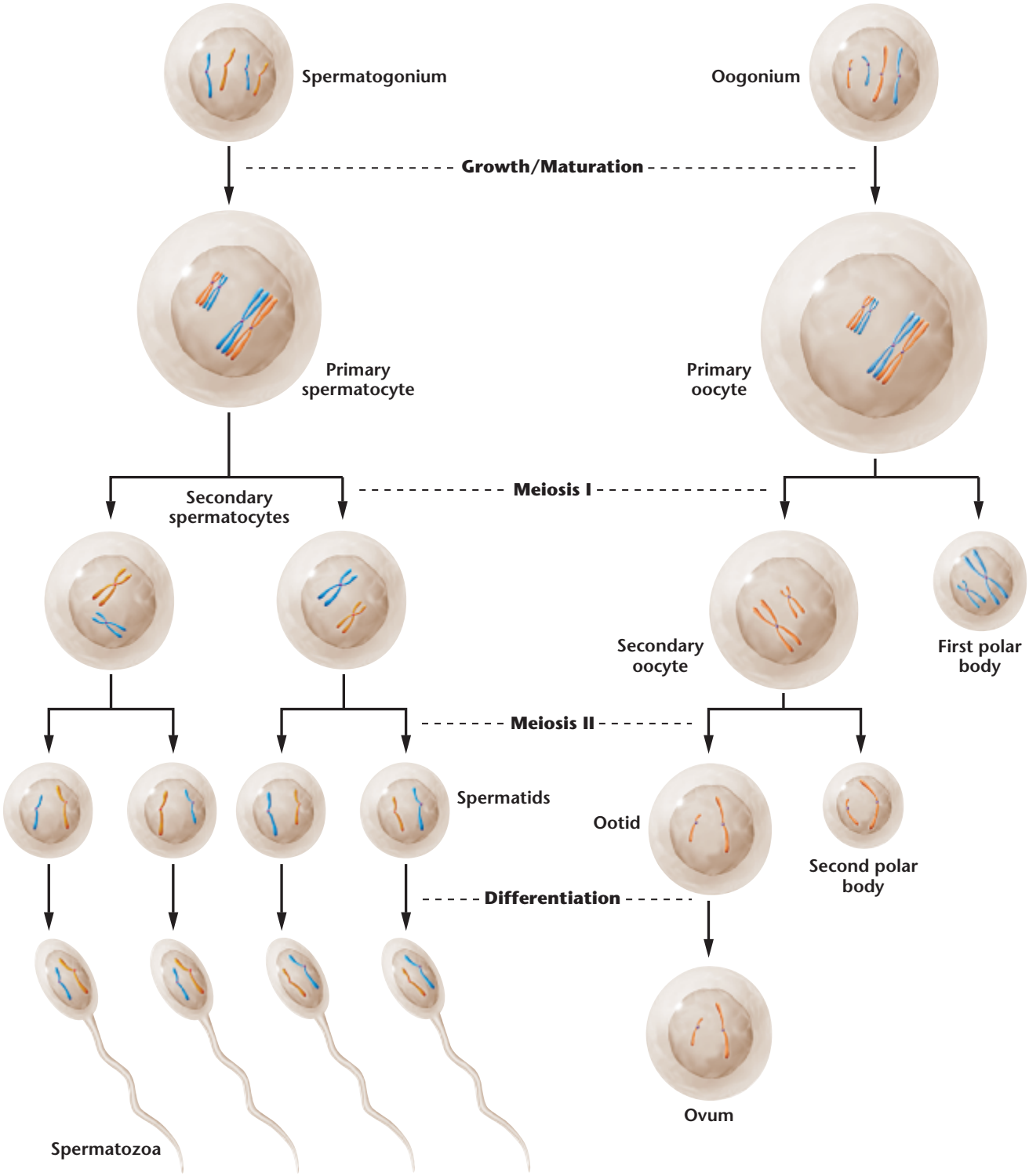


FIGURE 2.12 Spermatogenesis and oogenesis in animal cells.

2.5 The Development of Gametes Varies in Spermatogenesis Compared to Oogenesis

Although events that occur during the meiotic divisions are similar in all cells participating in gametogenesis in most animal species, there are certain differences between the production of a male gamete (spermatogenesis) and a female gamete (oogenesis). **Figure 2.12** summarizes these processes.

Spermatogenesis takes place in the testes, the male reproductive organs. The process begins with the enlargement of an undifferentiated diploid germ cell called a *spermatogonium.* This cell grows to become a *primary spermatocyte,* which undergoes the first meiotic division. The products of this division, called *secondary spermatocytes,* contain a haploid number of dyads. The secondary spermatocytes then undergo meiosis II, and each of these cells produces two haploid *spermatids.* Spermatids go through a series of developmental changes, *spermiogenesis,* to become highly specialized, motile *spermatozoa,* or *sperm.* All sperm cells produced during spermatogenesis contain the haploid number of chromosomes and equal amounts of cytoplasm.

Spermatogenesis may be continuous or may occur periodically in mature male animals; its onset is determined by the species' reproductive cycles. Animals that reproduce year-round produce sperm continuously, whereas those whose breeding period is confined to a particular season produce sperm only during that time.

In animal *oogenesis,* the formation of *ova* (sing. **ovum**), or eggs, occurs in the ovaries, the female reproductive organs. The daughter cells resulting from the two meiotic divisions of this process receive equal amounts of genetic material, but they do *not* receive equal amounts of cytoplasm. Instead, during each division, almost all the cytoplasm of the *primary oocyte,* itself derived from the *oogonium,* is concentrated in one of the two daughter cells. The concentration of cytoplasm is necessary because a major function of the mature ovum is to nourish the developing embryo following fertilization.

During anaphase I in oogenesis, the tetrads of the primary oocyte separate, and the dyads move toward opposite poles. During telophase I, the dyads at one pole are pinched off with very little surrounding cytoplasm to form the *first polar body.* The first polar body may or may not divide again to produce two small haploid cells. The other daughter cell produced by this first meiotic division contains most of the cytoplasm and is called the *secondary oocyte.* The mature ovum will be produced from the secondary oocyte during the second meiotic division. During this division, the cytoplasm of the secondary oocyte again divides unequally, producing an **ootid** and a **second polar body**. The ootid then differentiates into the mature ovum.

Unlike the divisions of spermatogenesis, the two meiotic divisions of oogenesis may not be continuous. In some animal species, the second division may directly follow the first. In others, including humans, the first division of all oocytes begins in the embryonic ovary but arrests in prophase I. Many years later, meiosis resumes in each oocyte just prior to its ovulation. The second division is completed only after fertilization.

NOW SOLVE THIS

2.3 Examine Figure 2.12, which shows oogenesis in animal cells. Will the genotype of the second polar body (derived from meiosis II) always be identical to that of the ootid? Why or why not?

■ **HINT:** *This problem involves an understanding of meiosis during oogenesis, asking you to demonstrate your knowledge of polar bodies. The key to its solution is to take into account that crossing over occurred between each pair of homologs during meiosis I.*

2.6 Meiosis Is Critical to Sexual Reproduction in All Diploid Organisms

The process of meiosis is critical to the successful sexual reproduction of all diploid organisms. It is the mechanism by which the diploid amount of genetic information is reduced to the haploid amount. In animals, meiosis leads to the formation of gametes, whereas in plants haploid spores are produced, which in turn lead to the formation of haploid gametes.

Each diploid organism stores its genetic information in the form of homologous pairs of chromosomes. Each pair consists of one member derived from the maternal parent and one from the paternal parent. Following meiosis, haploid cells potentially contain either the paternal or the maternal representative of every homologous pair of chromosomes. However, the process of crossing over, which occurs in the first meiotic prophase, further reshuffles the alleles between the maternal and paternal members of each homologous pair, which then segregate and assort independently into gametes. These events result in the great amount of genetic variation present in gametes.

It is important to touch briefly on the significant role that meiosis plays in the life cycles of fungi and plants. In many fungi, the predominant stage of the life cycle consists of haploid vegetative cells. They arise through meiosis and proliferate by mitotic cell division. In multicellular plants, the life cycle alternates between the diploid **sporophyte stage**

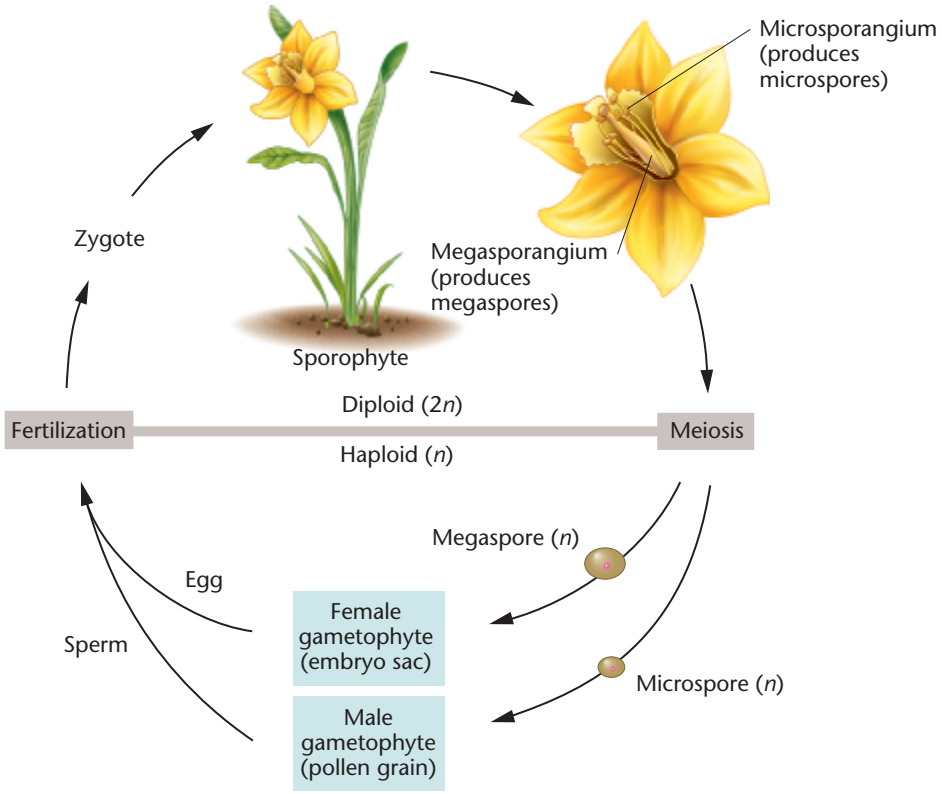


FIGURE 2.13 Alternation of generations between the diploid sporophyte (2*n*) and the haploid gametophyte (*n*) in a multicellular plant. The processes of meiosis and fertilization bridge the two phases of the life cycle. In angiosperms (flowering plants), like the one shown here, the sporophyte stage is the predominant phase.

and the haploid *gametophyte stage* (Figure 2.13). While one or the other predominates in different plant groups during this "alternation of generations," the processes of meiosis and fertilization constitute the "bridges" between the sporophyte and gametophyte stages. Therefore, meiosis is an essential component of the life cycle of plants.

2.7 Electron Microscopy Has Revealed the Physical Structure of Mitotic and Meiotic Chromosomes

Thus far in this chapter, we have focused on mitotic and meiotic chromosomes, emphasizing their behavior during cell division and gamete formation. An interesting question is why chromosomes are invisible during interphase but visible during the various stages of mitosis and meiosis. Studies using electron microscopy clearly show why this is the case.

Recall that, during interphase, only dispersed chromatin fibers are present in the nucleus [Figure 2.14(a)]. Once mitosis begins, however, the fibers coil and fold, condensing into typical mitotic chromosomes [Figure 2.14(b)]. If the fibers comprising a mitotic chromosome are loosened, the areas of greatest spreading reveal individual fibers similar to those seen in interphase chromatin [Figure 2.14(c)]. Very few fiber ends seem to be present, and in some cases, none can be seen. Instead, individual fibers always seem to loop back into the interior. Such fibers are obviously twisted and coiled around one another, forming the regular pattern of folding in the mitotic chromosome. Starting in late telophase of mitosis and continuing during G1 of interphase, chromosomes unwind to form the long fibers characteristic of chromatin, which consist of DNA and associated proteins, particularly proteins called *histones*. It is in this physical arrangement that DNA can most efficiently function during transcription and replication.

Electron microscopic observations of metaphase chromosomes in varying degrees of coiling led Ernest DuPraw to postulate the **folded-fiber model**, shown in Figure 2.14(c). During metaphase, each chromosome consists of two sister chromatids joined at the centromeric region. Each arm of the chromatid appears to be a single fiber wound much like a skein of yarn. The fiber is composed of tightly coiled double-stranded DNA and protein. An orderly coiling–twisting–condensing

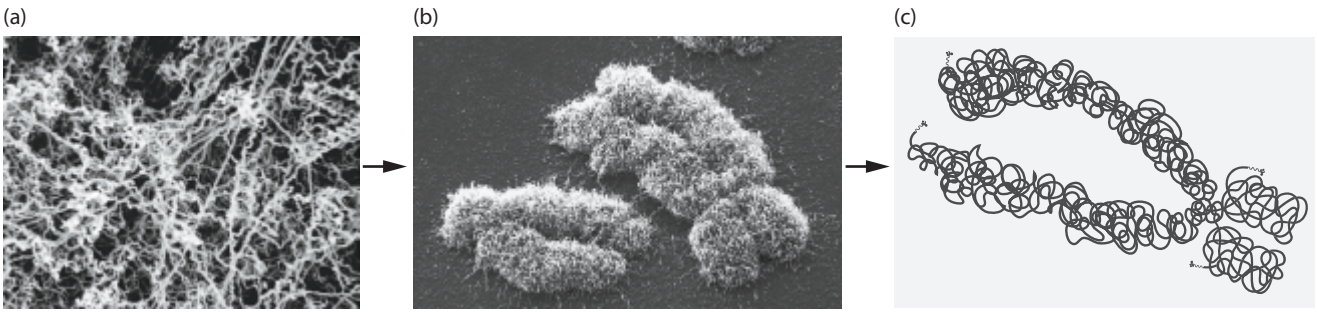


FIGURE 2.14 Comparison of (a) the chromatin fibers characteristic of the interphase nucleus with (b) metaphase chromosomes that are derived from chromatin during mitosis. Part (c) diagrams a mitotic chromosome, showing how chromatin is condensed to produce it. Part (a) is a transmission electron micrograph and part (b) is a scanning electron micrograph.

process appears to facilitate the transition of the interphase chromatin into the more condensed mitotic chromosomes. Geneticists believe that during the transition from interphase to prophase, a 5000-fold compaction occurs in the length of DNA within the chromatin fiber! This process must be extremely precise given the highly ordered and consistent appearance of mitotic chromosomes in all eukaryotes. Note particularly in the micrographs the clear distinction between the sister chromatids constituting each chromosome. They are joined only by the common centromere that they share prior to anaphase. We will return to this general topic later in the text when we consider chromosome structure in further detail (see Chapter 12).

EXPLORING GENOMICS

PubMed: Exploring and Retrieving Biomedical Literature

Mastering Genetics Visit the Study Area: Exploring Genomics

PubMed is an Internet-based search system developed by the National Center of Biotechnology Information (NCBI) at the National Library of Medicine. Using PubMed, one can access over 26 million citations for publications in over 5600 biomedical journals. The full text of many of the articles can be obtained electronically through college or university libraries, and some journals (such as *Proceedings of the National Academy of Sciences USA; Genome Biology;* and *Science*) provide free public access to articles within certain time frames.

In this exercise, we will explore PubMed to answer questions about relationships between tubulin, human cancers, and cancer therapies.

Exercise I – Tubulin, Cancer, and Mitosis

In this chapter we were introduced to tubulin and the dynamic behavior of microtubules during the cell cycle. Cancer cells are characterized by continuous and uncontrolled mitotic divisions.

Is it possible that tubulin and microtubules contribute to the development of cancer? Could these important structures be targets for cancer therapies?

1. To begin your search for the answers, access the PubMed site at http://www.ncbi.nlm.nih.gov/pubmed/.
2. In the search box, type "tubulin cancer" and then click the "Search" button to perform the search.
3. Select several research papers and read the abstracts.

To answer the question about tubulin's association with cancer, you may want to limit your search to fewer papers, perhaps those that are review articles. To do this, click the "Review" link under the Article Types category on the left side of the page.

Explore some of the articles, as abstracts or as full text, available in your library or by free public access. Prepare a brief report or verbally share your experiences with your class. Describe two of the most important things you learned during your exploration, and identify the information sources you encountered during the search.

CASE STUDY Timing is everything

Over a period of two years, a man in his early 20s received a series of intermittent chemotherapy and radiotherapy treatments for Hodgkin disease. During this therapy, he and his wife were unable to initiate a pregnancy. The man had a series of his semen samples examined at a fertility clinic. The findings revealed that shortly after each treatment very few mature sperm were present, and abnormal chromosome numbers were often observed in developing spermatocytes. However, such chromosome abnormalities disappeared about 40 days after treatment, and normal sperm reappeared about 74 days post-treatment.

1. How might a genetic counselor explain the time-related differences in sperm production and the appearance and subsequent disappearance of chromosomal abnormalities?
2. Do you think that exposure to chemotherapy and radiotherapy would cause more problems to spermatocytes than to mature sperm?
3. Prior to treatment, should the physician(s) involved have been ethically obligated to recommend genetic counseling? What advice regarding fertility might have been suggested?

For further reading, see: Harel, S., et al., 2011. Management of fertility in patients treated for Hodgkin's lymphoma. *Haematologica* 2011. 96: 1692–99.

Summary Points

Mastering Genetics For activities, animations, and review quizzes, go to the Study Area.

1. The structure of cells is elaborate and complex, with most components involved directly or indirectly with genetic processes.
2. In diploid organisms, chromosomes exist in homologous pairs, where each member is identical in size, centromere placement, and gene loci. One member of each pair is derived from the maternal parent, and the other from the paternal parent.
3. Mitosis and meiosis are mechanisms by which cells distribute the genetic information contained in their chromosomes to progeny cells in a precise, orderly fashion.
4. Mitosis, which is but one part of the cell cycle, is subdivided into discrete stages that initially depict the condensation of chromatin into the diploid number of chromosomes. Each chromosome first appears as a double structure, consisting of a pair of identical sister chromatids joined at a common centromere. As mitosis proceeds, centromeres split and sister chromatids are pulled apart by spindle fibers and directed toward opposite poles of the cell. Cytoplasmic division then occurs, creating two new cells with the identical genetic information contained in the progenitor cell.
5. Meiosis converts a diploid cell into haploid gametes or spores, making sexual reproduction possible. As a result of chromosome duplication, two subsequent meiotic divisions are required to achieve haploidy, whereby each haploid cell receives one member of each homologous pair of chromosomes.
6. There is a major difference between meiosis in males and in females. Spermatogenesis partitions the cytoplasmic volume equally and produces four haploid sperm cells. Oogenesis, on the other hand, collects the bulk of cytoplasm in one egg cell and reduces the other haploid products to polar bodies. The extra cytoplasm in the egg contributes to zygote development following fertilization.
7. Meiosis results in extensive genetic variation by virtue of the exchange of chromosome segments during crossing over between maternal and paternal chromatids and by virtue of the random separation of maternal and paternal chromatids into gametes. In addition, meiosis plays an important role in the life cycles of fungi and plants, serving as the bridge between alternating generations.
8. Mitotic chromosomes are produced as a result of the coiling and condensation of chromatin fibers of interphase into the characteristic form of chromatids.

INSIGHTS AND SOLUTIONS

This appearance of "Insights and Solutions" begins a feature that will have great value to you as a student. From this point on, "Insights and Solutions" precedes the "Problems and Discussion Questions" at each chapter's end to provide sample problems and solutions that demonstrate approaches you will find useful in genetic analysis. The insights you gain by working through the sample problems will improve your ability to solve the ensuing problems in each chapter.

1. In an organism with a diploid number of $2n = 6$, how many individual chromosomal structures will align on the metaphase plate during (a) mitosis, (b) meiosis I, and (c) meiosis II? Describe each configuration.

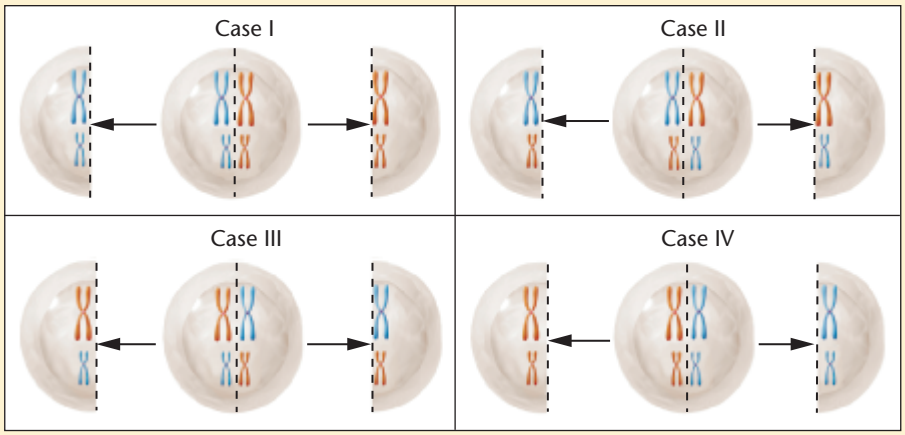
Solution: (a) Remember that in mitosis, homologous chromosomes do not synapse, so there will be six double structures, each consisting of a pair of sister chromatids. In other words, the number of structures is equivalent to the diploid number.

(b) In meiosis I, the homologs have synapsed, reducing the number of structures to three. Each is called a tetrad and consists of two pairs of sister chromatids.

(c) In meiosis II, the same number of structures exist (three), but in this case they are called dyads. Each dyad is a pair of sister chromatids. When crossing over has occurred, each chromatid may contain parts of one of its nonsister chromatids, obtained during exchange in prophase I.

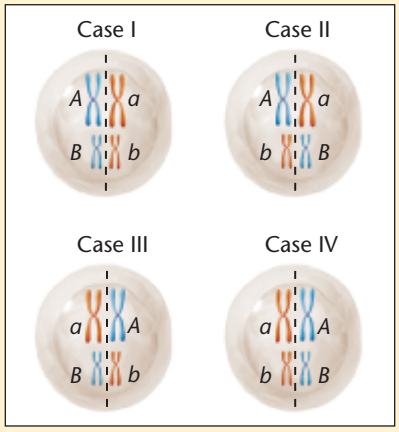
2. Disregarding crossing over, draw all possible alignment configurations that can occur during metaphase for the chromosomes shown in Figure 2.11.

Solution: As shown in the diagram below, four configurations are possible when $n = 2$.



3. For the chromosomes in Problem 2, assume that each of the larger chromosomes has a different allele for a given gene, *A* OR *a*, as shown. Also assume that each of the smaller chromosomes has a different allele for a second gene, *B* OR *b*. Calculate the probability of generating each possible combination of these alleles (*AB*, *Ab*, *aB*, *ab*) following meiosis I.

Solution: As shown in the accompanying diagram:



Case I	*AB* and *ab*	**Total:** $AB = 2(p = 1/4)$
Case II	*Ab* and *aB*	$Ab = 2(p = 1/4)$
Case III	*aB* and *Ab*	$aB = 2(p = 1/4)$
Case IV	*ab* and *AB*	$ab = 2(p = 1/4)$

4. How many different chromosome configurations can occur following meiosis I if three different pairs of chromosomes are present ($n = 3$)?

Solution: If $n = 3$, then eight different configurations would be possible. The formula 2^n, where n equals the haploid number, represents the number of potential alignment patterns. As we will see in (Chapter 3), these patterns are produced according to the Mendelian postulate of *segregation,* and they serve as the physical basis of another Mendelian postulate called *independent assortment.*

5. Describe the composition of a meiotic tetrad during prophase I, assuming no crossover event has occurred. What impact would a single crossover event have on this structure?

Solution: Such a tetrad contains four chromatids, existing as two pairs. Members of each pair are sister chromatids. They are held together by a common centromere. Members of one pair are maternally derived, whereas members of the other are paternally derived. Maternal and paternal members are called nonsister chromatids. A single crossover event has the effect of exchanging a portion of a maternal and a paternal chromatid, leading to a chiasma, where the two involved chromatids overlap physically in the tetrad. The process of exchange is referred to as crossing over.

Problems and Discussion Questions

Mastering Genetics Visit for instructor-assigned tutorials and problems.

1. **HOW DO WE KNOW?** In this chapter, we focused on how chromosomes are distributed during cell division, both in dividing somatic cells (mitosis) and in gamete- and spore-forming cells (meiosis). We found many opportunities to consider the methods and reasoning by which much of this information was acquired. From the explanations given in the chapter, answer the following questions.
(a) How do we know that chromosomes exist in homologous pairs?
(b) How do we know that DNA replication occurs during interphase, not early in mitosis?
(c) How do we know that mitotic chromosomes are derived from chromatin?
2. **CONCEPT QUESTION** Review the Chapter Concepts list on page 50. All of these pertain to conceptual issues involving mitosis or meiosis. Based on these concepts, write a short essay that contrasts mitosis and meiosis, including their respective roles in organisms, the mechanisms by which they achieve their respective outcomes, and the consequences should either process fail to be executed with absolute fidelity.
3. What role do the following cellular components play in the storage, expression, or transmission of genetic information: (a) chromatin, (b) nucleolus, (c) ribosome, (d) mitochondrion, (e) centriole, (f) centromere?
4. Discuss the concepts of homologous chromosomes, diploidy, and haploidy. What characteristics do two homologous chromosomes share?
5. If two chromosomes of a species are the same length and have similar centromere placements and yet are not homologous, what is different about them?
6. Describe the events that characterize each stage of mitosis.
7. How do spindle fibers form and how do chromosomes separate in animal cells?
8. Compare chromosomal separation in plant and animal cells.
9. Why might different cells of the same organism have cell cycles of different durations?
10. Define and discuss these terms: (a) synapsis, (b) bivalents, (c) chiasmata, (d) crossing over, (e) chromomeres, (f) sister chromatids, (g) tetrads, (h) dyads, (i) monads.
11. A diploid organism has alleles *T* and *t* for the same gene on a pair of homologous chromosomes. In what circumstances might both alleles segregate? At what stage of cell division would this occur?
12. Given the end results of the two types of division, why is it necessary for homologs to pair during meiosis and not desirable for them to pair during mitosis?
13. Contrast spermatogenesis and oogenesis. What is the significance of the formation of polar bodies?
14. How do the stages of mitosis and meiosis occur in a specific order and never alternate?
15. A diploid cell contains three pairs of homologous chromosomes designated C1 and C2, M1 and M2, and S1 and S2. No crossing over occurs. What combinations of chromosomes are possible in (a) daughter cells following mitosis, (b) cells undergoing the first meiotic metaphase, (c) haploid cells following both divisions of meiosis?
16. Predict the number of unique haploid gametes that could be produced through meiosis in an organism with a diploid number of $2n = 16$. Assume that crossing over does not occur.
17. During oogenesis in an animal species with a haploid number of 6, one dyad undergoes nondisjunction during meiosis II. Following the second meiotic division, this dyad ends up intact in the ovum. How many chromosomes are present in (a) the mature ovum and (b) the second polar body? (c) Following fertilization by a normal sperm, what chromosome condition is created?
18. Humans have a diploid number of 46. What is the probability that a sperm will be formed that contains all chromosomes whose centromeres are derived from maternal homologs?
19. Cattle (*Bos taurus*) have a diploid number of 60, and their haploid DNA content per cell is approximately 3.2 picogram. What would be the DNA content of a somatic cell (non-sex cell) at anaphase? What would be the nuclear DNA content of a secondary spermatocyte? What would be the nuclear DNA content of a spermatozoon?
20. Describe the role of meiosis in the life cycle of a vascular plant.
21. How many sister chromatids are seen in the metaphase for a single chromosome? How different are these structures from the interphase chromatin?
22. What is the significance of checkpoints in the cell cycle?
23. A metaphase chromosome preparation from an unknown organism clearly shows 40 chromosomes that can be easily paired. However, there are two unmatched chromosomes that differ from each other in size and centromere placement. What can you say about the chromosomes and the ploidy of this organism?
24. If one follows 50 primary oocytes in an animal through their various stages of oogenesis, how many secondary oocytes would be formed? How many first polar bodies would be formed? How many ootids would be formed? If one follows 50 primary spermatocytes in an animal through their various stages of spermatogenesis, how many secondary spermatocytes would be formed? How many spermatids would be formed?

Extra-Spicy Problems

Mastering Genetics Visit for instructor-assigned tutorials and problems.

As part of the "Problems and Discussion Questions" section in this and each subsequent chapter, we shall present a number of "Extra-Spicy" genetics problems. We have chosen to set these apart in order to identify problems that are particularly challenging. You may be asked to examine and assess actual data, to design genetics experiments, or to engage in cooperative learning. Like genetic varieties of peppers, some of these experiences are just spicy and some are very hot. We hope that you will enjoy the challenges that they pose.

For Problems **25–30**, consider a diploid cell that contains three pairs of chromosomes designated AA, BB, and CC. Each pair contains a maternal and a paternal member (e.g., A^m and A^p). Using these designations, demonstrate your understanding of mitosis and meiosis by drawing chromatid combinations as requested. Be sure to indicate when chromatids are paired as a result of replication and/or synapsis. You may wish to use a large piece of brown manila wrapping paper or a cut-up paper grocery bag for this project and to work in partnership

with another student. We recommend cooperative learning as an efficacious way to develop the skills you will need for solving the problems presented throughout this text.

25. In mitosis, what chromatid combination(s) will be present during metaphase? What combination(s) will be present at each pole at the completion of anaphase?

26. During meiosis I, assuming no crossing over, what chromatid combination(s) will be present at the completion of prophase I? Draw all possible alignments of chromatids as migration begins during early anaphase.

27. Are there any possible combinations present during prophase of meiosis II other than those that you drew in Problem 26? If so, draw them.

28. Draw all possible combinations of chromatids during the early phases of anaphase in meiosis II.

29. Assume that during meiosis I none of the *C* chromosomes disjoin at metaphase, but they separate into dyads (instead of monads) during meiosis II. How would this change the alignments that you constructed during the anaphase stages in meiosis I and II? Draw them.

30. Assume that each gamete resulting from Problem 29 fuses, in fertilization, with a normal haploid gamete. What combinations will result? What percentage of zygotes will be diploid, containing one paternal and one maternal member of each chromosome pair?

31. A species of cereal rye (*Secale cereale*) has a chromosome number of 14, while a species of Canadian wild rye (*Elymus canadensis*) has a chromosome number of 28. Sterile hybrids can be produced by crossing *Secale* with *Elymus*.
(a) What would be the expected chromosome number in the somatic cells of the hybrids?
(b) Given that none of the chromosomes pair at meiosis I in the sterile hybrid (Hang and Franckowlak, 1984), speculate on the anaphase I separation patterns of these chromosomes.

32. An interesting procedure has been applied for assessing the chromosomal balance of potential secondary oocytes for use in human *in vitro* fertilization. Using fluorescence *in situ* hybridization (FISH), Kuliev and Verlinsky (2004) were able to identify individual chromosomes in first polar bodies and thereby infer the chromosomal makeup of "sister" oocytes. Assume that when examining a first polar body you saw that it had one copy (dyad) of each chromosome but two dyads of chromosome 21. What would you expect to be the chromosomal 21 complement in the secondary oocyte? What consequences are likely in the resulting zygote, if the secondary oocyte was fertilized?

33. Assume that you were examining a first polar body and noted that it had one copy (dyad) of each chromosome except chromosome 21. Chromosome 21 was completely absent. What would you expect to be the chromosome 21 complement (only with respect to chromosome 21) in the secondary oocyte? What consequences are likely in the resulting zygote if the secondary oocyte was fertilized?

34. Kuliev and Verlinsky (2004) state that there was a relatively high number of separation errors at meiosis I. In these cases the centromere underwent a premature division, occurring at meiosis I rather than meiosis II. Regarding chromosome 21, what would you expect to be the chromosome 21 complement in the secondary oocyte in which you saw a single chromatid (monad) for chromosome 21 in the first polar body? If this secondary oocyte was involved in fertilization, what would be the expected consequences?

3

Mendelian Genetics

Gregor Johann Mendel, who in 1866 put forward the major postulates of transmission genetics as a result of experiments with the garden pea.

CHAPTER CONCEPTS

- Inheritance is governed by information stored in discrete unit factors called genes.
- Genes are transmitted from generation to generation on vehicles called chromosomes.
- Chromosomes, which exist in pairs in diploid organisms, provide the basis of biparental inheritance.
- During gamete formation, chromosomes are distributed according to postulates first described by Gregor Mendel, based on his nineteenth-century research with the garden pea.
- Mendelian postulates prescribe that homologous chromosomes segregate from one another and assort independently with other segregating homologs during gamete formation.
- Genetic ratios, expressed as probabilities, are subject to chance deviation and may be evaluated statistically.
- The analysis of pedigrees allows predictions concerning the genetic nature of human traits.

Although inheritance of biological traits has been recognized for thousands of years, the first significant insights into how it takes place only occurred about 150 years ago. In 1866, Gregor Johann Mendel published the results of a series of experiments that would lay the foundation for the formal discipline of genetics. Mendel's work went largely unnoticed until the turn of the twentieth century, but eventually, the concept of the gene as a distinct hereditary unit was established. Since then, the ways in which genes, as segments of chromosomes, are transmitted to offspring and control traits have been clarified. Research continued unabated throughout the twentieth century and into the present—indeed, studies in genetics, most recently at the molecular level, have remained at the forefront of biological research since the early 1900s.

When Mendel began his studies of inheritance using *Pisum sativum,* the garden pea, chromosomes and the role and mechanism of meiosis were totally unknown. Nevertheless, he determined that discrete units of inheritance exist and predicted their behavior in the formation of gametes. Subsequent investigators, with access to cytological data, were able to relate their own observations of chromosome behavior during meiosis and Mendel's principles of inheritance. Once this correlation was recognized, Mendel's postulates were accepted as the basis for the study of what is known as **transmission genetics**—how genes are transmitted from parents to offspring. These principles were derived directly from Mendel's experimentation.

3.1 Mendel Used a Model Experimental Approach to Study Patterns of Inheritance

Johann Mendel was born in 1822 to a peasant family in the Central European village of Heinzendorf. An excellent student in high school, he studied philosophy for several years afterward and in 1843, taking the name Gregor, was admitted to the Augustinian Monastery of St. Thomas in Brno, now part of the Czech Republic. In 1849, he was relieved of pastoral duties, and from 1851 to 1853, he attended the University of Vienna, where he studied physics and botany. He returned to Brno in 1854, where he taught physics and natural science for the next 16 years. Mendel received support from the monastery for his studies and research throughout his life.

In 1856, Mendel performed his first set of hybridization experiments with the garden pea, launching the research phase of his career. His experiments continued until 1868, when he was elected abbot of the monastery. Although he retained his interest in genetics, his new responsibilities demanded most of his time. In 1884, Mendel died of a kidney disorder. The local newspaper paid him the following tribute:

> His death deprives the poor of a benefactor, and mankind at large of a man of the noblest character, one who was a warm friend, a promoter of the natural sciences, and an exemplary priest.

Mendel first reported the results of some simple genetic crosses between certain strains of the garden pea in 1865. Although his was not the first attempt to provide experimental evidence pertaining to inheritance, Mendel's success where others had failed can be attributed, at least in part, to his elegant experimental design and analysis.

Mendel showed remarkable insight into the methodology necessary for good experimental biology. First, he chose an organism that was easy to grow and to hybridize artificially. The pea plant is self-fertilizing in nature, but it is easy to cross-breed experimentally. It reproduces well and grows to maturity in a single season. Mendel followed seven visible features (we refer to them as characters, or characteristics), each represented by two contrasting forms, or **traits** (**Figure 3.1**). For the character stem height, for example, he experimented with the traits *tall* and *dwarf*. He selected

Character	Contrasting traits		F_1 results	F_2 results	F_2 ratio
Seed shape	round/wrinkled		all round	5474 round 1850 wrinkled	2.96:1
Seed color	yellow/green		all yellow	6022 yellow 2001 green	3.01:1
Pod shape	full/constricted		all full	882 full 299 constricted	2.95:1
Pod color	green/yellow		all green	428 green 152 yellow	2.82:1
Flower color	violet/white		all violet	705 violet 224 white	3.15:1
Flower position	axial/terminal		all axial	651 axial 207 terminal	3.14:1
Stem height	tall/dwarf		all tall	787 tall 277 dwarf	2.84:1

FIGURE 3.1 Seven pairs of contrasting traits and the results of Mendel's seven monohybrid crosses of the garden pea (*Pisum sativum*). In each case, pollen derived from plants exhibiting one trait was used to fertilize the ova of plants exhibiting the other trait. In the F_1 generation, one of the two traits was exhibited by all plants. The contrasting trait reappeared in approximately 1/4 of the F_2 plants.

six other contrasting pairs of traits involving seed shape and color, pod shape and color, and flower color and position. From local seed merchants, Mendel obtained true-breeding strains, those in which each trait appeared unchanged generation after generation in self-fertilizing plants.

There were several other reasons for Mendel's success. In addition to his choice of a suitable organism, he restricted his examination to one or very few pairs of contrasting traits in each experiment. He also kept accurate quantitative records, a necessity in genetic experiments. From the analysis of his data, Mendel derived certain postulates that have become the principles of transmission genetics.

The results of Mendel's experiments went unappreciated until the turn of the century, well after his death. However, once Mendel's publications were rediscovered by geneticists investigating the function and behavior of chromosomes, the implications of his postulates were immediately apparent. He had discovered the basis for the transmission of hereditary traits!

3.2 The Monohybrid Cross Reveals How One Trait Is Transmitted from Generation to Generation

Mendel's simplest crosses involved only one pair of contrasting traits. Each such experiment is called a **monohybrid cross**. A monohybrid cross is made by mating true-breeding individuals from two parent strains, each exhibiting one of the two contrasting forms of the character under study. Initially, we examine the first generation of offspring of such a cross, and then we consider the offspring of **selfing**, that is, of self-fertilization of individuals from this first generation. The original parents constitute the $\mathbf{P_1}$, or **parental generation**; their offspring are the F_1, or **first filial generation**; the individuals resulting from the selfed F_1 generation are the $\mathbf{F_2}$, or **second filial generation**; and so on.

The cross between true-breeding pea plants with tall stems and dwarf stems is representative of Mendel's monohybrid crosses. *Tall* and *dwarf* are contrasting traits of the character of stem height. Unless tall or dwarf plants are crossed together or with another strain, they will undergo self-fertilization and breed true, producing their respective traits generation after generation. However, when Mendel crossed tall plants with dwarf plants, the resulting F_1 generation consisted of only tall plants. When members of the F_1 generation were selfed, Mendel observed that 787 of 1064 F_2 plants were tall, while 277 of 1064 were dwarf. Note that in this cross (Figure 3.1), the dwarf trait disappeared in the F_1 generation, only to reappear in the F_2 generation.

Genetic data are usually expressed and analyzed as ratios. In this particular example, many identical P_1 crosses were made and many F_1 plants—all tall—were produced. As noted, of the 1064 F_2 offspring, 787 were tall and 277 were dwarf—a ratio of approximately 2.8:1.0, or about 3:1.

Mendel made similar crosses between pea plants exhibiting each of the other pairs of contrasting traits; the results of these crosses are shown in Figure 3.1. In every case, the outcome was similar to the tall/dwarf cross just described. For the character of interest, all F_1 offspring expressed the same trait exhibited by one of the parents, but in the F_2 offspring, an approximate ratio of 3:1 was obtained. That is, three-fourths looked like the F_1 plants, while one-fourth exhibited the contrasting trait, which had disappeared in the F_1 generation.

We note one further aspect of Mendel's monohybrid crosses. In each cross, the F_1 and F_2 patterns of inheritance were similar regardless of which P_1 plant served as the source of pollen (sperm) and which served as the source of the ovum (egg). The crosses could be made either way—pollination of dwarf plants by tall plants, or vice versa. Crosses made in both these ways are called **reciprocal crosses**. Therefore, the results of Mendel's monohybrid crosses were not sex dependent.

To explain these results, Mendel proposed the existence of particulate *unit factors* for each trait. He suggested that these factors serve as the basic units of heredity and are passed unchanged from generation to generation, determining various traits expressed by each individual plant. Using these general ideas, Mendel proceeded to hypothesize precisely how such factors could account for the results of the monohybrid crosses.

Mendel's First Three Postulates

Using the consistent pattern of results in the monohybrid crosses, Mendel derived the following three postulates, or principles, of inheritance.

1. **UNIT FACTORS IN PAIRS**
   Genetic characters are controlled by unit factors existing in pairs in individual organisms.
   In the monohybrid cross involving tall and dwarf stems, a specific **unit factor** exists for each trait. Each diploid individual receives one factor from each parent. Because the factors occur in pairs, three combinations are possible: two factors for tall stems, two factors for dwarf stems, or one of each factor. Every individual possesses one of these three combinations, which determines stem height.

2. **DOMINANCE/RECESSIVENESS**
   When two unlike unit factors responsible for a single character are present in a single individual, one unit factor is dominant to the other, which is said to be recessive.
   In each monohybrid cross, the trait expressed in the F_1 generation is controlled by the dominant unit factor. The trait not expressed is controlled by the recessive unit factor. The terms dominant and recessive are also

used to designate traits. In this case, tall stems are said to be dominant over recessive dwarf stems.

3. **SEGREGATION**
   During the formation of gametes, the paired unit factors separate, or segregate, randomly so that each gamete receives one or the other with equal likelihood.
   If an individual contains a pair of like unit factors (e.g., both specific for tall), then all its gametes receive one of that same kind of unit factor (in this case, tall). If an individual contains unlike unit factors (e.g., one for tall and one for dwarf), then each gamete has a 50 percent probability of receiving either the tall or the dwarf unit factor.

These postulates provide a suitable explanation for the results of the monohybrid crosses. Let's use the tall/dwarf cross to illustrate. Mendel reasoned that P_1 tall plants contained identical paired unit factors, as did the P_1 dwarf plants. The gametes of tall plants all receive one tall unit factor as a result of segregation. Similarly, the gametes of dwarf plants all receive one dwarf unit factor. Following fertilization, all F_1 plants receive one unit factor from each parent—a tall factor from one and a dwarf factor from the other—reestablishing the paired relationship, but because tall is dominant to dwarf, all F_1 plants are tall.

When F_1 plants form gametes, the postulate of segregation demands that each gamete randomly receives either the tall *or* dwarf unit factor. Following random fertilization events during F_1 selfing, four F_2 combinations will result with equal frequency:

1. tall/tall
2. tall/dwarf
3. dwarf/tall
4. dwarf/dwarf

Combinations (1) and (4) will clearly result in tall and dwarf plants, respectively. According to the postulate of dominance/recessiveness, combinations (2) and (3) will both yield tall plants. Therefore, the F_2 is predicted to consist of 3/4 tall and 1/4 dwarf, or a ratio of 3:1. This is approximately what Mendel observed in his cross between tall and dwarf plants. A similar pattern was observed in each of the other monohybrid crosses (Figure 3.1).

Modern Genetic Terminology

To analyze the monohybrid cross and Mendel's first three postulates, we must first introduce several new terms as well as a symbol convention for the unit factors. Traits such as tall or dwarf are physical expressions of the information contained in unit factors. The physical expression of a trait is the **phenotype** of the individual. Mendel's unit factors represent units of inheritance called **genes** by modern geneticists. For any given character, such as plant height, the phenotype is determined by alternative forms of a single gene, called **alleles**. For example, the unit factors representing tall and dwarf are alleles determining the height of the pea plant.

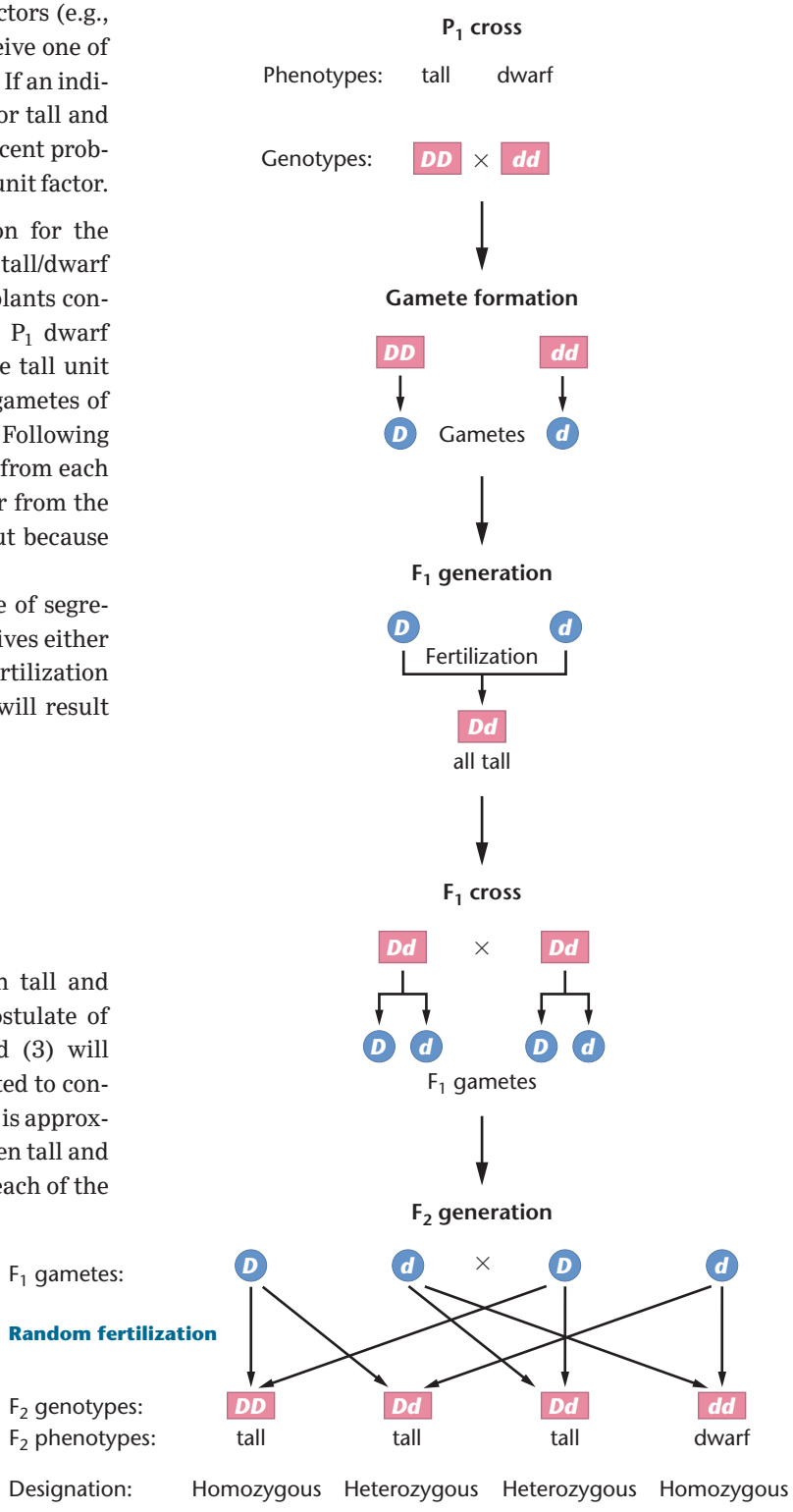


FIGURE 3.2 The monohybrid cross between tall (*D*) and dwarf (*d*) pea plants. Individuals are shown in rectangles, and gametes are shown in circles.

Geneticists have several different systems for using symbols to represent genes. Later in the text (see Chapter 4), we will review a number of these conventions, but for now, we will adopt one to use consistently throughout this chapter. According to this convention, the first letter of the recessive trait symbolizes the character in question; in lowercase italic, it designates the allele for the recessive trait, and in uppercase italic, it designates the allele for the dominant trait. Thus for Mendel's pea plants, we use *d* for the *d*warf allele and *D* for the tall allele. When alleles are written in pairs to represent the two unit factors present in any individual (*DD*, *Dd*, or *dd*), the resulting symbol is called the **genotype**. The genotype designates the genetic makeup of an individual for the trait or traits it describes, whether the individual is haploid or diploid. By reading the genotype, we know the phenotype of the individual: *DD* and *Dd* are tall, and *dd* is dwarf. When both alleles are the same (*DD* or *dd*), the individual is **homozygous** for the trait, or a **homozygote**; when the alleles are different (*Dd*), we use the terms **heterozygous** and **heterozygote**. These symbols and terms are used in **Figure 3.2** to describe the monohybrid cross, as discussed on page 75.

Punnett Squares

The genotypes and phenotypes resulting from combining gametes during fertilization can be easily visualized by constructing a diagram called a **Punnett square**, named after the person who first devised this approach, Reginald C. Punnett. **Figure 3.3** illustrates this method of analysis for our $F_1 \times F_1$ monohybrid cross. Each of the possible gametes is assigned a column or a row; the vertical columns represent those of the female parent, and the horizontal rows represent those of the male parent. After assigning the gametes to the rows and columns, we predict the new generation by entering the male and female gametic information into each box and thus producing every possible resulting genotype. By filling out the Punnett square, we are listing all possible random fertilization events. The genotypes and phenotypes of all potential offspring are ascertained by reading the combinations in the boxes.

The Punnett square method is particularly useful when you are first learning about genetics and how to solve genetics problems. Note the ease with which the 3:1 phenotypic ratio and the 1:2:1 genotypic ratio may be derived for the F_2 generation in Figure 3.3.

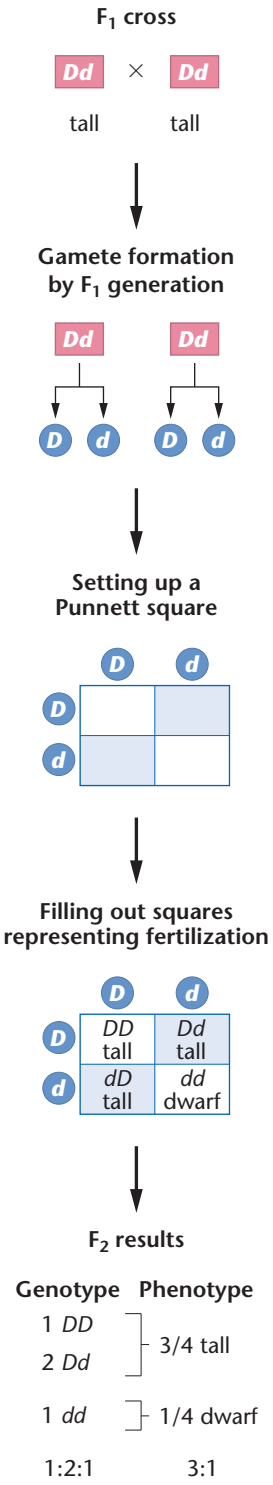


FIGURE 3.3 A Punnett square generating the F_2 ratio of the $F_1 \times F_1$ cross shown in Figure 3.2.

The Testcross: One Character

Tall plants produced in the F_2 generation are predicted to have either the *DD* or the *Dd* genotype. You might ask if there is a way to distinguish the genotype. Mendel devised a rather simple method that is still used today to discover the genotype of plants and animals: the **testcross**. The organism expressing the dominant phenotype but having an unknown genotype is crossed with a known *homozygous recessive individual*. For example, as shown in **Figure 3.4(a)**, if a tall plant of genotype *DD* is testcrossed with a dwarf plant, which must have the *dd* genotype, all offspring will be tall phenotypically and *Dd* genotypically. However, as shown in **Figure 3.4(b)**, if a tall plant is *Dd* and is crossed with a dwarf plant (*dd*), then one-half

NOW SOLVE THIS

3.1 Pigeons may exhibit a checkered or plain color pattern. In a series of controlled matings, the following data were obtained.

	F_1 Progeny	
P_1 Cross	Checkered	Plain
(a) checkered × checkered	36	0
(b) checkered × plain	38	0
(c) plain × plain	0	35

Then F_1 offspring were selectively mated with the following results. (The P_1 cross giving rise to each F_1 pigeon is indicated in parentheses.)

	F_2 Progeny	
F_1 × F_1 Crosses	Checkered	Plain
(d) checkered (a) × plain (c)	34	0
(e) checkered (b) × plain (c)	17	14
(f) checkered (b) × checkered (b)	28	9
(g) checkered (a) × checkered (b)	39	0

How are the checkered and plain patterns inherited? Select and assign symbols for the genes involved, and determine the genotypes of the parents and offspring in each cross.

■ **HINT:** *This problem asks you to analyze the data produced from several crosses involving pigeons and to determine the mode of inheritance and the genotypes of the parents and offspring in a number of instances. The key to its solution is to first determine whether or not this is a monohybrid cross. To do so, convert the data to ratios that are characteristic of Mendelian crosses. In the case of this problem, ask first whether any of the F_2 ratios match Mendel's 3:1 monohybrid ratio. If so, the second step is to determine which trait is dominant and which is recessive.*

of the offspring will be tall (*Dd*) and the other half will be dwarf (*dd*). Therefore, a 1:1 tall/dwarf ratio demonstrates the heterozygous nature of the tall plant of unknown genotype. The results of the testcross reinforced Mendel's conclusion that separate unit factors control traits.

3.3 Mendel's Dihybrid Cross Generated a Unique F_2 Ratio

As a natural extension of the monohybrid cross, Mendel also designed experiments in which he examined two characters simultaneously. Such a cross, involving two pairs of contrasting traits, is a **dihybrid cross**, or a *two-factor cross*. For example, if pea plants having yellow seeds that are round were bred with those having green seeds that are wrinkled, the results shown in Figure 3.5 would occur: the

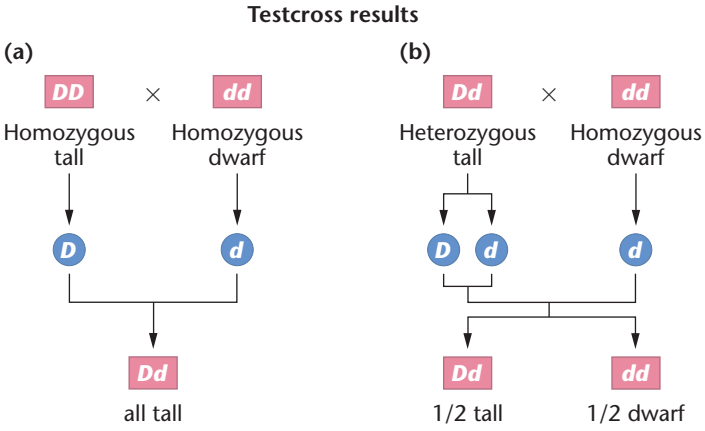


FIGURE 3.4 Testcross of a single character. In (a), the tall parent is homozygous, but in (b), the tall parent is heterozygous. The genotype of each tall P_1 plant can be determined by examining the offspring when each is crossed with the homozygous recessive dwarf plant.

F_1 offspring would all be yellow and round. It is therefore apparent that yellow is dominant to green and that round is dominant to wrinkled. When the F_1 individuals are selfed, approximately 9/16 of the F_2 plants express the yellow and round traits, 3/16 express yellow and wrinkled, 3/16 express green and round, and 1/16 express green and wrinkled.

A variation of this cross is also shown in Figure 3.5. Instead of crossing one P_1 parent with both dominant traits (yellow, round) to one with both recessive traits (green, wrinkled), plants with yellow, wrinkled seeds are crossed with those with green, round seeds. In spite of the change in the P_1 phenotypes, both the F_1 and F_2 results remain unchanged. Why this is so will become clear below.

Mendel's Fourth Postulate: Independent Assortment

We can most easily understand the results of a dihybrid cross if we consider it theoretically as consisting of two monohybrid crosses conducted separately. Think of the two sets of traits as being inherited independently of each other; that is, the chance of any plant having yellow or green seeds is not at all influenced by the chance that this plant will have round or wrinkled seeds. Thus, because yellow is dominant to green, all F_1 plants in the first theoretical cross would have yellow seeds. In the second theoretical cross, all F_1 plants would have round seeds because round is dominant to wrinkled. When Mendel examined the F_1 plants of the dihybrid cross, all were yellow and round, as our theoretical crosses predict.

The predicted F_2 results of the first cross are 3/4 yellow and 1/4 green. Similarly, the second cross would yield 3/4 round and 1/4 wrinkled. Figure 3.5 shows that in the dihybrid cross, 12/16 F_2 plants are yellow, while 4/16 are green, exhibiting the expected 3:1 (3/4:1/4) ratio. Similarly, 12/16 of all F_2 plants have round seeds, while 4/16 have wrinkled seeds, again revealing the 3:1 ratio.

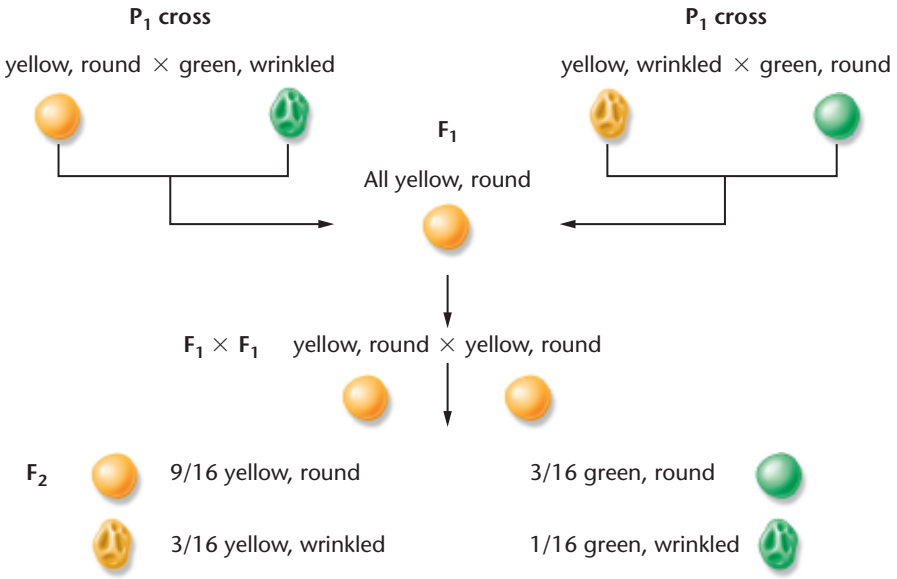


FIGURE 3.5 F_1 and F_2 results of Mendel's dihybrid crosses in which the plants on the top left with yellow, round seeds are crossed with plants having green, wrinkled seeds, and the plants on the top right with yellow, wrinkled seeds are crossed with plants having green, round seeds.

These numbers demonstrate that the two pairs of contrasting traits are inherited independently, so we can predict the frequencies of all possible F_2 phenotypes by applying the **product law** of probabilities: *the probability of two or more independent events occurring simultaneously is equal to the product of their individual probabilities.* For example, the probability of an F_2 plant having yellow and round seeds is (3/4)(3/4), or 9/16, because 3/4 of all F_2 plants should be yellow and 3/4 of all F_2 plants should be round.

In a like manner, the probabilities of the other three F_2 phenotypes can be calculated: yellow (3/4) and wrinkled (1/4) are predicted to be present together 3/16 of the time; green (1/4) and round (3/4) are predicted 3/16 of the time; and green (1/4) and wrinkled (1/4) are predicted 1/16 of the time. These calculations are shown in **Figure 3.6**.

It is now apparent why the F_1 and F_2 results are identical whether the initial cross is yellow, round plants bred with green, wrinkled plants, or whether yellow, wrinkled plants are bred with green, round plants. In both crosses, the F_1 genotype of all offspring is identical. As a result, the F_2 generation is also identical in both crosses.

On the basis of similar results in numerous dihybrid crosses, Mendel proposed a fourth postulate:

4. INDEPENDENT ASSORTMENT
   During gamete formation, segregating pairs of unit factors assort independently of each other.
   This postulate stipulates that segregation of any pair of unit factors occurs independently of all others. As a result of random segregation, each gamete receives one member of every pair of unit factors. For one pair, whichever unit factor is received does not influence the outcome of segregation of any other pair. Thus, according to the postulate of independent assortment, all possible combinations of gametes should be formed in equal frequency.

The Punnett square in **Figure 3.7** shows how independent assortment works in the formation of the F_2 generation. Examine the formation of gametes by the F_1 plants; segregation prescribes that every gamete receives either a *G* or *g* allele and a *W* or *w* allele. Independent assortment stipulates that all four combinations (*GW*, *Gw*, *gW*, and *gw*) will be formed with equal probabilities.

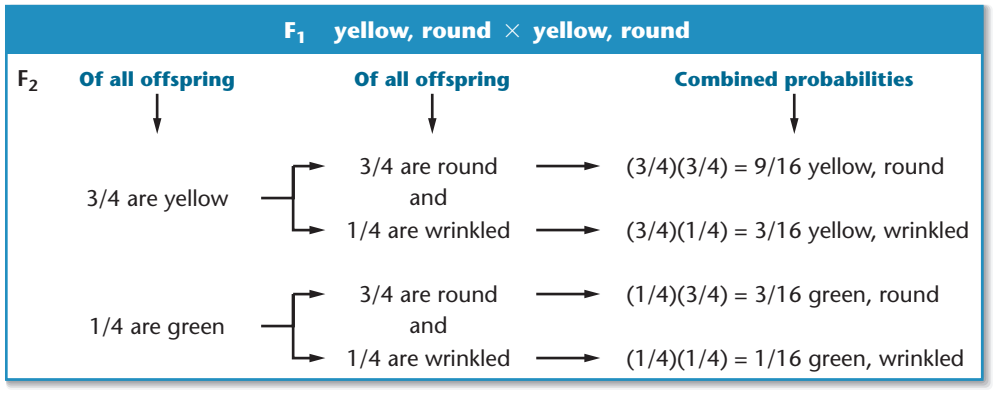


FIGURE 3.6 Computation of the combined probabilities of each F_2 phenotype for two independently inherited characters. The probability of each plant being yellow or green is independent of the probability of it bearing round or wrinkled seeds.

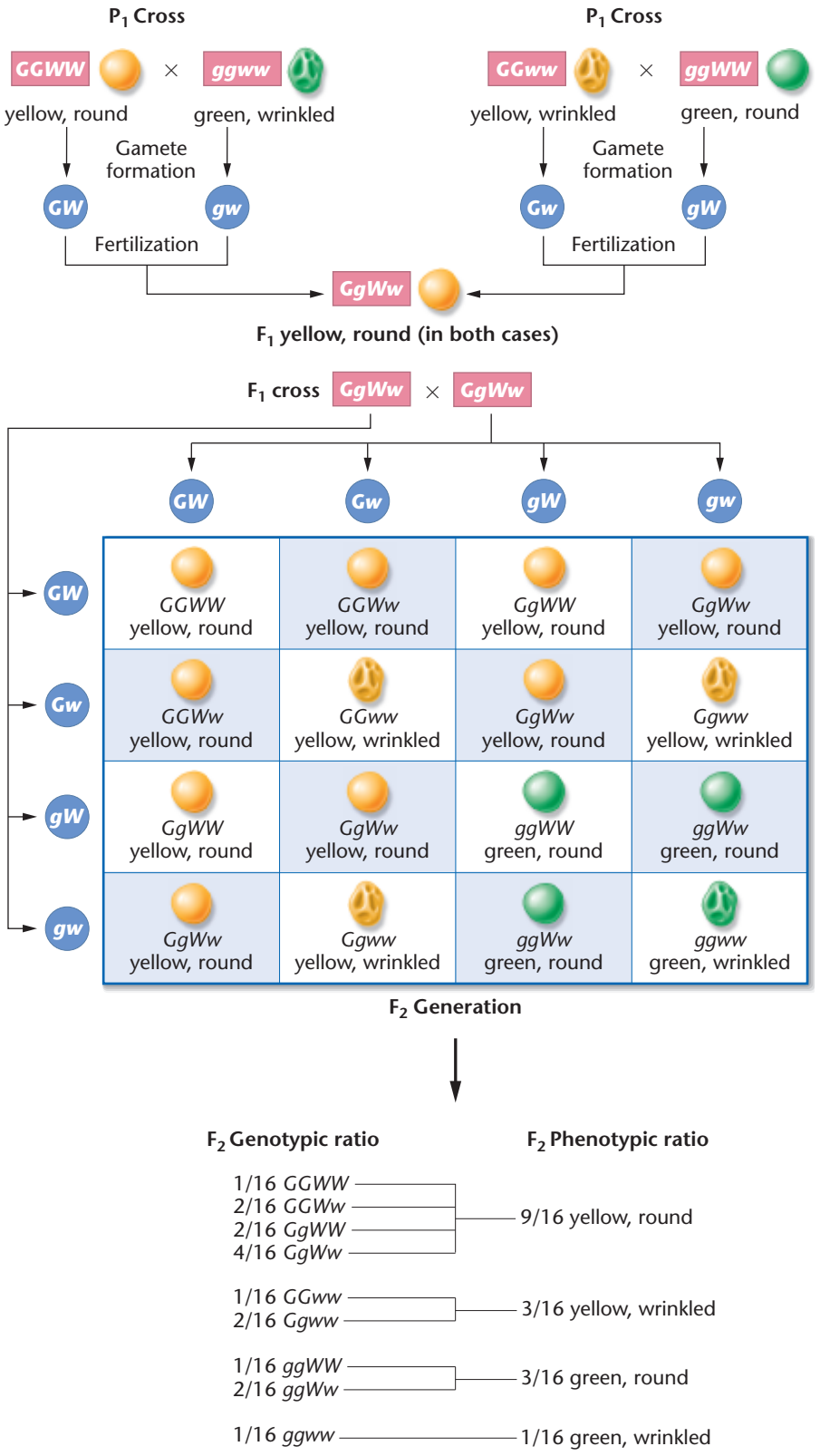


FIGURE 3.7 Analysis of the dihybrid crosses shown in Figure 3.5. The F_1 heterozygous plants are self-fertilized to produce an F_2 generation, which is computed using a Punnett square. Both the phenotypic and genotypic F_2 ratios are shown.

MODERN APPROACHES TO UNDERSTANDING GENE FUNCTION

Identifying Mendel's Gene for Regulating White Flower Color in Peas

In 2010, 150 years after Gregor Mendel studied pea flower color, an international team of researchers identified the gene responsible for regulating flower color in peas. The potential gene they focused on was called pea gene *A*. This gene was also found in other plants, including petunias and barrel clovers. Gene *A* encodes a protein that functions as a **transcription factor**—a protein that binds to DNA and regulates expression of other genes. Cells in purple flowers of pea plants accumulate anthocyanin pigment molecules that are responsible for their color. Pea plants with white flowers do not accumulate anthocyanin, even though they contain the gene that encodes the enzyme involved in anthocyanin synthesis. Researchers hypothesized that the transcription factor produced by pea gene *A* might regulate expression of the anthocyanin biosynthetic gene.

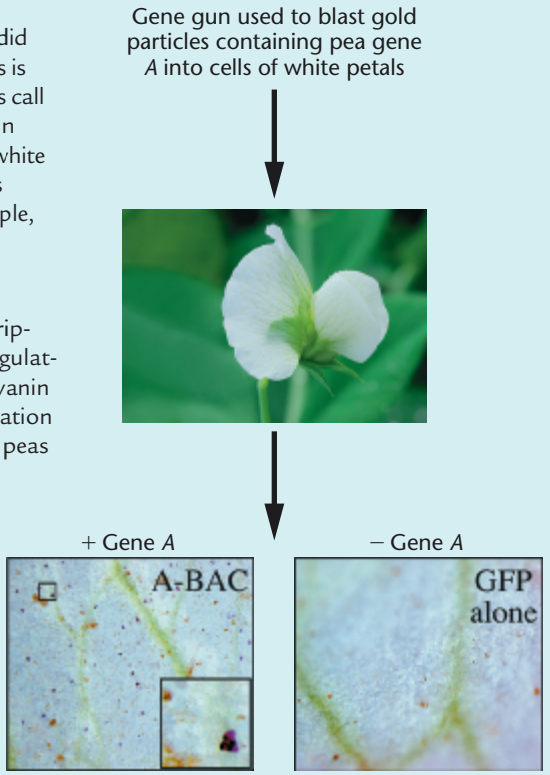
To test this hypothesis and confirm gene *A* function, they delivered normal copies of gene *A* into white flower petals by using a gene gun, a device that shoots metal particles coated with a gene of interest into cells. In this approach, gold particles coated with gene *A* enter a small percentage of cells and gene *A* is expressed in those cells.

Results:

Cells of white petals where gene *A* is expressed (left photo) accumulate anthocyanin pigment and turn purple. The inset square shows a higher magnification image enlarging spots of gene *A* expression. A control experiment where white petals were treated with DNA without gene *A* (right photo) did not restore pigmentation. This is an example of what geneticists call a **rescue experiment** because in cells that received gene *A* the white flower mutant phenotype was rescued or restored to the purple, wild-type phenotype.

Conclusion:

Pea gene *A* encodes a transcription factor responsible for regulating expression of the anthocyanin gene in peas. Further examination of the gene *A* sequence from peas with white flowers revealed a single-nucleotide change in gene *A* that renders this transcription factor inactive. Cells with a normal copy of gene *A* express anthocyanin and turn purple. Cells with a mutant form of gene *A* do not accumulate anthocyanin and are white. The genetic mystery of Mendel's white flowers had been solved.



Reference:

Hellens, R. P., et al. (2010). Identification of Mendel's White Flower Character. *PLoS One* 5(10): e13230. doi:10.1371/journal.pone.0013230.

Take a look at the YouTube video "Finding the molecular answer to Mendel's pea colour experiments" to hear Dr. Hellens describe the approach his group used to identify the function of this gene. http://www.youtube.com/watch?v=BEhtyXCdcTg

Questions to Consider:

1. Why do you think that expression of gene *A* appears as spots in the leaves shown in the photo on the left? What does this signify?
2. If you did the same experiment with a pea plant that had a mutation in the gene for anthocyanin accumulation, would you expect that introduction of gene *A* would rescue the phenotype of the mutant? Why or why not?

In every $F_1 \times F_1$ fertilization event, each zygote has an equal probability of receiving one of the four combinations from each parent. If many offspring are produced, 9/16 have yellow, round seeds, 3/16 have yellow, wrinkled seeds, 3/16 have green, round seeds, and 1/16 have green, wrinkled seeds, yielding what is designated as **Mendel's 9:3:3:1 dihybrid ratio**. This is an ideal ratio based on probability events involving segregation, independent assortment, and random fertilization. Because of deviation due strictly to chance, particularly if small numbers of offspring are produced, actual results are highly unlikely to match the ideal ratio.

The Testcross: Two Characters

The testcross may also be applied to individuals that express two dominant traits but whose genotypes are unknown. For example, the expression of the yellow, round seed phenotype in the F_2 generation just described may result from the *GGWW, GGWw, GgWW*, or *GgWw* genotypes. If an F_2 yellow,

NOW SOLVE THIS

3.2 Considering the Mendelian traits round versus wrinkled and yellow versus green, consider the crosses below and determine the genotypes of the parental plants by analyzing the phenotypes of their offspring.

Parental Plants	Offspring
(a) round, yellow × round, yellow	3/4 round, yellow 1/4 wrinkled, yellow
(b) wrinkled, yellow × round, yellow	6/16 wrinkled, yellow 2/16 wrinkled, green 6/16 round, yellow 2/16 round, green
(c) round, yellow × round, yellow	9/16 round, yellow 3/16 round, green 3/16 wrinkled, yellow 1/16 wrinkled, green
(d) round, yellow × wrinkled, green	1/4 round, yellow 1/4 round, green 1/4 wrinkled, yellow 1/4 wrinkled, green

■ **HINT:** *This problem involves a series of Mendelian dihybrid crosses where you are asked to determine the genotypes of the parents in a number of instances. The key to its solution is to write down everything that you know for certain. This reduces the problem to its bare essentials, clarifying what you need to determine. For example, the wrinkled, yellow plant in case (b) must be homozygous for the recessive wrinkled alleles and bear at least one dominant allele for the yellow trait. Having established this, you need only determine the remaining allele for seed color.*

round plant is crossed with the homozygous recessive green, wrinkled plant (*ggww*), analysis of the offspring will indicate the exact genotype of that yellow, round plant. Each of the above genotypes results in a different set of gametes and, in a testcross, a different set of phenotypes in the resulting offspring. You should work out the results of each of these four crosses to be sure that you understand this concept.

3.4 The Trihybrid Cross Demonstrates That Mendel's Principles Apply to Inheritance of Multiple Traits

Thus far, we have considered inheritance of up to two pairs of contrasting traits. Mendel demonstrated that the processes of segregation and independent assortment also apply to three pairs of contrasting traits, in what is called a **trihybrid cross**, or *three-factor cross.*

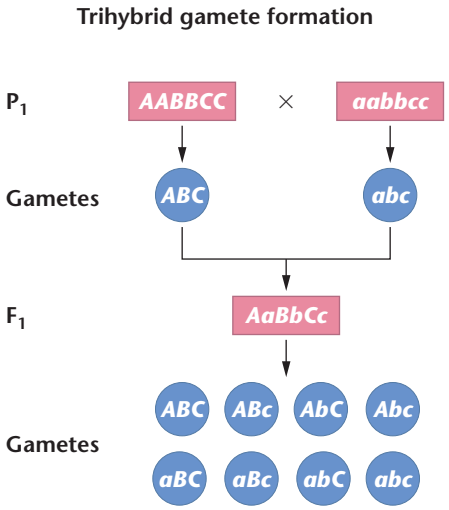


FIGURE 3.8 Formation of P_1 and F_1 gametes in a trihybrid cross.

Although a trihybrid cross is somewhat more complex than a dihybrid cross, its results are easily calculated if the principles of segregation and independent assortment are followed. For example, consider the cross shown in **Figure 3.8** where the allele pairs of theoretical contrasting traits are represented by the symbols *A, a, B, b, C,* and *c*. In the cross between *AABBCC* and *aabbcc* individuals, all F_1 individuals are heterozygous for all three gene pairs. Their genotype, *AaBbCc*, results in the phenotypic expression of the dominant *A, B,* and *C* traits. When F_1 individuals serve as parents, each produces eight different gametes in equal frequencies. At this point, we could construct a Punnett square with 64 separate boxes and read out the phenotypes—but such a method is cumbersome in a cross involving so many factors. Therefore, another method has been devised to calculate the predicted ratio.

The Forked-Line Method, or Branch Diagram

It is much less difficult to consider each contrasting pair of traits separately and then to combine these results by using the **forked-line method**, first shown in Figure 3.6. This method, also called a **branch diagram**, relies on the simple application of the laws of probability established for the dihybrid cross. Each gene pair is assumed to behave independently during gamete formation.

When the monohybrid cross $Aa \times aa$ is made, we know that:

1. All F_1 individuals have the genotype *Aa* and express the phenotype represented by the *A* allele, which is called the *A* phenotype in the discussion that follows.
2. The F_2 generation consists of individuals with either the *A* phenotype or the *a* phenotype in the ratio of 3:1.

Generation of F_2 trihybrid phenotypes

A or *a*	*B* or *b*	C or *c*	Combined proportion
3/4 *A*	3/4 *B*	3/4 C	(3/4)(3/4)(3/4) *ABC* = 27/64 *ABC*
		1/4 *c*	(3/4)(3/4)(1/4) *ABc* = 9/64 *ABc*
	1/4 *b*	3/4 C	(3/4)(1/4)(3/4) *AbC* = 9/64 *AbC*
		1/4 *c*	(3/4)(1/4)(1/4) *Abc* = 3/64 *Abc*
1/4 *a*	3/4 *B*	3/4 C	(1/4)(3/4)(3/4) *aBC* = 9/64 *aBC*
		1/4 *c*	(1/4)(3/4)(1/4) *aBc* = 3/64 *aBc*
	1/4 *b*	3/4 C	(1/4)(1/4)(3/4) *abC* = 3/64 *abC*
		1/4 *c*	(1/4)(1/4)(1/4) *abc* = 1/64 *abc*

FIGURE 3.9 Generation of the F_2 trihybrid phenotypic ratio using the forked-line method. This method is based on the expected probability of occurrence of each phenotype.

The same generalizations can be made for the *BB* × *bb* and *CC* × *cc* crosses. Thus, in the F_2 generation, 3/4 of all organisms will express phenotype *A*, 3/4 will express *B*, and 3/4 will express *C*. Similarly, 1/4 of all organisms will express *a*, 1/4 will express *b*, and 1/4 will express *c*. The proportions of organisms that express each phenotypic combination can be predicted by assuming that fertilization, following the independent assortment of these three gene pairs during gamete formation, is a random process. We apply the product law of probabilities once again. **Figure 3.9** uses the forked-line method to calculate the phenotypic proportions of the F_2 generation. They fall into the trihybrid ratio of 27:9:9:9:3:3:3:1. The same method can be used to solve crosses involving any number of gene pairs, *provided that all gene pairs assort independently from each other.* We shall see later that gene pairs do not always assort with complete independence. However, it appeared to be true for all of Mendel's characters.

NOW SOLVE THIS

3.3 Using the forked-line, or branch diagram, method, determine the genotypic and phenotypic ratios of these trihybrid crosses: (a) *AaBbCc* × *AaBBCC*, (b) *AaBBCc* × *aaBBCc*, and (c) *AaBbCc* × *AaBbCc*.

■ **HINT:** *This problem asks you to use the forked-line method to determine the outcome of a number of trihybrid crosses. The key to its solution is to realize that in using the forked-line method, you must consider each gene pair separately. For example, in this problem, first predict the outcome of each cross for the A /a genes, then for the B/b genes, and finally, for the C/c genes. Then you are prepared to pursue the outcome of each cross using the forked-line method.*

3.5 Mendel's Work Was Rediscovered in the Early Twentieth Century

Mendel published his work in 1866. While his findings were often cited and discussed, their significance went unappreciated for about 35 years. Then, in the latter part of the nineteenth century, a remarkable observation set the scene for the recognition of Mendel's work: Walter Flemming's discovery of chromosomes in the nuclei of salamander cells. In 1879, Flemming described the behavior of these thread-like structures during cell division. As a result of his findings and the work of many other cytologists, the presence of discrete units within the nucleus soon became an integral part of scientists' ideas about inheritance.

In the early twentieth century, hybridization experiments similar to Mendel's were performed independently by three botanists, Hugo de Vries, Carl Correns, and Erich Tschermak. De Vries's work demonstrated the principle of segregation in several plant species. Apparently, he searched the existing literature and found that Mendel's work had anticipated his own conclusions! Correns and Tschermak also reached conclusions similar to those of Mendel.

About the same time, two cytologists, Walter Sutton and Theodor Boveri, independently published papers linking their discoveries of the behavior of chromosomes during meiosis to the Mendelian principles of segregation and independent assortment. They pointed out that the separation of chromosomes during meiosis could serve as the cytological basis of these two postulates. Although they thought that Mendel's unit factors were probably chromosomes rather than genes on chromosomes, their findings reestablished the importance of Mendel's work and led to many ensuing genetic investigations. Sutton and Boveri are credited with initiating the **chromosomal theory of inheritance**, the idea that the genetic material in living organisms is contained in chromosomes, which was developed during the next two decades. As we will see in subsequent chapters, work by Thomas H. Morgan, Alfred H. Sturtevant, Calvin Bridges, and others established beyond a reasonable doubt that Sutton's and Boveri's hypothesis was correct.

Unit Factors, Genes, and Homologous Chromosomes

Because the correlation between Sutton's and Boveri's observations and Mendelian postulates serves as the foundation for the modern description of transmission genetics, we will examine this correlation in some depth before moving on to other topics.

As we know, each species possesses a specific number of chromosomes in each somatic cell nucleus. For diploid organisms, this number is called the **diploid number (2*n*)** and is characteristic of that species. During the formation of gametes (meiosis), the number is precisely halved (*n*), and when two gametes combine during fertilization, the diploid number is reestablished. During meiosis, however, the chromosome number is not reduced in a random manner. It was apparent to early cytologists that the diploid number of chromosomes is composed of homologous pairs identifiable by their morphological appearance and behavior. The gametes contain one member of each pair—thus the chromosome complement of a gamete is quite specific, and the number of chromosomes in each gamete is equal to the haploid number.

With this basic information, we can see the correlation between the behavior of unit factors and chromosomes and genes. Figure 3.10 shows three of Mendel's postulates and

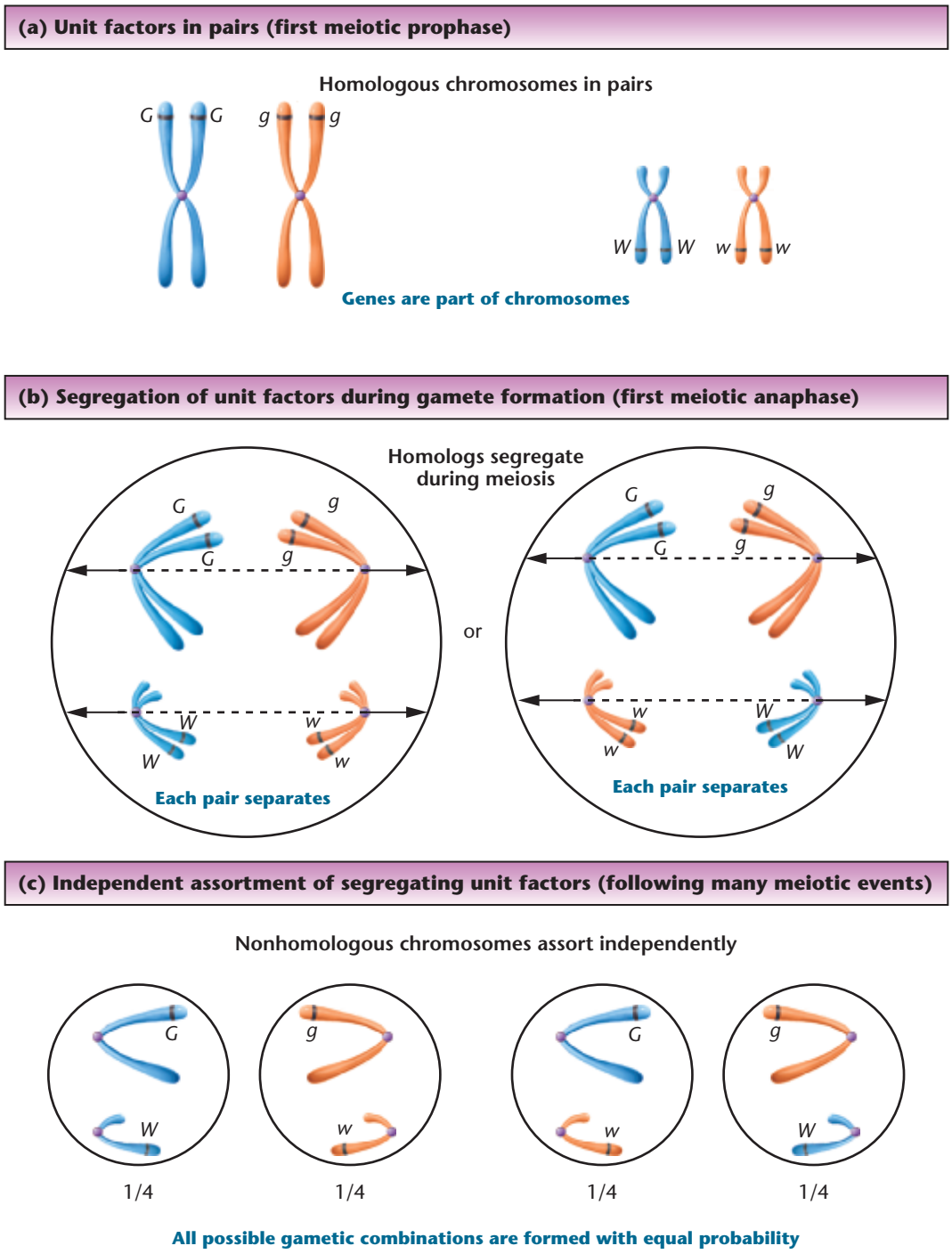


FIGURE 3.10 Illustrated correlation between the Mendelian postulates of (a) unit factors in pairs, (b) segregation, and (c) independent assortment, showing the presence of genes located on homologous chromosomes and their behavior during meiosis.

the chromosomal explanation of each. Unit factors are really genes located on homologous pairs of chromosomes [Figure 3.10(a)]. Members of each pair of homologs separate, or segregate, during gamete formation [Figure 3.10(b)]. In the figure, two different alignments are possible, both of which are shown.

To illustrate the principle of independent assortment, it is important to distinguish between members of any given homologous pair of chromosomes. One member of each pair is derived from the **maternal parent**, whereas the other comes from the **paternal parent**. (We represent the different parental origins with different colors.) As shown in Figure 3.10(c), following independent segregation of each pair of homologs, each gamete receives one member from each pair of chromosomes. All possible combinations are formed with equal probability. If we add the symbols used in Mendel's dihybrid cross (*G, g* and *W, w*) to the diagram, we can see why equal numbers of the four types of gametes are formed. The independent behavior of Mendel's pairs of unit factors (*G* and *W* in this example) is due to their presence on separate pairs of homologous chromosomes.

Observations of the phenotypic diversity of living organisms make it logical to assume that there are many more genes than chromosomes. Therefore, each homolog must carry genetic information for more than one trait. The currently accepted concept is that a chromosome is composed of a large number of linearly ordered, information-containing genes. Mendel's paired unit factors (which determine tall or dwarf stems, for example) actually constitute a pair of genes located on one pair of homologous chromosomes. The location on a given chromosome where any particular gene occurs is called its **locus** (pl. loci). The different alleles of a given gene (for example, *G* and *g*) contain slightly different genetic information (green or yellow) that determines the same character (seed color in this case). Although we have examined only genes with two alternative alleles, most genes have more than two allelic forms. We conclude this section by reviewing the criteria necessary to classify two chromosomes as a homologous pair:

1. During mitosis and meiosis, when chromosomes are visible in their characteristic shapes, both members of a homologous pair are the same size and exhibit identical centromere locations. The sex chromosomes (e.g., the X and the Y chromosomes in mammals) are an exception.
2. During early stages of meiosis, homologous chromosomes form pairs, or synapse.
3. Although it is not generally visible under the microscope, homologs contain the identical linear order of gene loci.

EVOLVING CONCEPT OF THE GENE

Based on the pioneering work of Gregor Mendel, the gene was viewed as a heritable unit factor that determines the expression of an observable trait, or phenotype. ■

3.6 Independent Assortment Leads to Extensive Genetic Variation

One consequence of independent assortment is the production by an individual of genetically dissimilar gametes. Genetic variation results because the two members of any homologous pair of chromosomes are rarely, if ever, genetically identical. As the maternal and paternal members of all pairs are distributed to gametes through independent assortment, all possible chromosome combinations are produced, leading to extensive genetic diversity.

We have seen that the number of possible gametes, each with different chromosome compositions, is 2^n, where n equals the haploid number. Thus, if a species has a haploid number of 4, then 2^4, or 16, different gamete combinations can be formed as a result of independent assortment. Although this number is not high, consider the human species, where $n = 23$. When 2^{23} is calculated, we find that in excess of 8×10^6, or over 8 million, different types of gametes are possible through independent assortment. Because fertilization represents an event involving only one of approximately 8×10^6 possible gametes from each of two parents, each offspring represents only one of $(8 \times 10^6)^2$ or one of only 64×10^{12} potential genetic combinations. Given that this probability is less than one in one trillion, it is no wonder that, except for identical twins, each member of the human species exhibits a distinctive set of traits—this number of combinations of chromosomes is far greater than the number of humans who have ever lived on Earth! Genetic variation resulting from independent assortment has been extremely important to the process of evolution in all sexually reproducing organisms.

3.7 Laws of Probability Help to Explain Genetic Events

Recall that genetic ratios—for example, 3/4 tall:1/4 dwarf—are most properly thought of as probabilities. These values predict the outcome of each fertilization event, such that the probability of each zygote having the genetic potential for becoming tall is 3/4, whereas the potential for its being a dwarf is 1/4. Probabilities range from 0.0, where an event *is certain not to occur,* to 1.0, where an event *is certain to occur.* In this section, we consider the relation of probability to genetics. When two

or more events with known probabilities occur independently but at the same time, we can calculate the probability of their possible outcomes occurring together. This is accomplished by applying the **product law**, which states that *the probability of two or more independent events occurring simultaneously is equal to the product of their individual probabilities* (see Section 3.3). Two or more events are independent of one another if the outcome of each one does not affect the outcome of any of the others under consideration.

To illustrate the product law, consider the possible results if you toss a penny (*P*) and a nickel (*N*) at the same time and examine all combinations of heads (*H*) and tails (*T*) that can occur. There are four possible outcomes:

$$(P_H{:}N_H) = (1/2)(1/2) = 1/4$$
$$(P_T{:}N_H) = (1/2)(1/2) = 1/4$$
$$(P_H{:}N_T) = (1/2)(1/2) = 1/4$$
$$(P_T{:}N_T) = (1/2)(1/2) = 1/4$$

The probability of obtaining a head or a tail in the toss of either coin is 1/2 and is unrelated to the outcome for the other coin. Thus, all four possible combinations are predicted to occur with equal probability.

If we want to calculate the probability when the possible outcomes of two events are independent of one another but can be accomplished in more than one way, we can apply the **sum law**. For example, what is the probability of tossing our penny and nickel and obtaining one head and one tail? In such a case, we do not care whether it is the penny or the nickel that comes up heads, provided that the other coin has the alternative outcome. As we saw above, there are two ways in which the desired outcome can be accomplished, each with a probability of 1/4. The sum law states that *the probability of obtaining any single outcome, where that outcome can be achieved by two or more events, is equal to the sum of the individual probabilities of all such events.* Thus, according to the sum law, the overall probability in our example is equal to

$$(1/4) + (1/4) = 1/2$$

One-half of all two-coin tosses are predicted to yield the desired outcome.

These simple probability laws will be useful throughout our discussions of transmission genetics and for solving genetics problems. In fact, we already applied the product law when we used the forked-line method to calculate the phenotypic results of Mendel's dihybrid and trihybrid crosses. When we wish to know the results of a cross, we need only calculate the probability of each possible outcome. The results of this calculation then allow us to predict the proportion of offspring expressing each phenotype or each genotype.

An important point to remember when you deal with probability is that predictions of possible outcomes are based on large sample sizes. If we predict that 9/16 of the offspring of a dihybrid cross will express both dominant traits, it is very unlikely that, in a small sample, exactly 9 of every 16 will express this phenotype. Instead, our prediction is that, of a large number of offspring, approximately 9/16 will do so. The deviation from the predicted ratio in smaller sample sizes is attributed to chance, a subject we examine in our discussion of statistics in Section 3.8. As you shall see, the impact of deviation due strictly to chance diminishes as the sample size increases.

3.8 Chi-Square Analysis Evaluates the Influence of Chance on Genetic Data

Mendel's 3:1 monohybrid and 9:3:3:1 dihybrid ratios are hypothetical predictions based on the following assumptions: (1) each allele is dominant or recessive, (2) segregation is unimpeded, (3) independent assortment occurs, and (4) fertilization is random. The final two assumptions are influenced by chance events and therefore are subject to random fluctuation. This concept of **chance deviation** is most easily illustrated by tossing a single coin numerous times and recording the number of heads and tails observed. In each toss, there is a probability of 1/2 that a head will occur and a probability of 1/2 that a tail will occur. Therefore, the expected ratio of many tosses is 1/2:1/2, or 1:1. If a coin is tossed 1000 times, usually *about* 500 heads and 500 tails will be observed. Any reasonable fluctuation from this hypothetical ratio (e.g., 486 heads and 514 tails) is attributed to chance.

As the total number of tosses is reduced, the impact of chance deviation increases. For example, if a coin is tossed only four times, you would not be too surprised if all four tosses resulted in only heads or only tails. For 1000 tosses, however, 1000 heads or 1000 tails would be most unexpected. In fact, you might believe that such a result would be impossible. Actually, all heads or all tails in 1000 tosses can be predicted to occur with a probability of $(1/2)^{1000}$. Since $(1/2)^{20}$ is less than one in a million times, an event occurring with a probability as small as $(1/2)^{1000}$ is virtually impossible. Two major points to keep in mind when predicting or analyzing genetic outcomes are:

1. The outcomes of independent assortment and fertilization, like coin tossing, are subject to random fluctuations from their predicted occurrences as a result of chance deviation.
2. As the sample size increases, the average deviation from the expected results decreases. Therefore, a larger sample size diminishes the impact of chance deviation on the final outcome.