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Antibodies
A Laboratory Manual, Second Edition

Edited by Edward A. Greenfield, Dana-Farber Cancer Institute

The second edition of the now-classic lab manual Antibodies, by Harlow and Lane, has been revised, extended, and updated by Edward Greenfield of the Dana-Farber Cancer Center, with contributions from other leaders in the field. This manual continues to be an essential resource for molecular biology, immunology, and cell culture labs on all matters relating to antibodies. The chapters on hybridomas and monoclonal antibodies have been recast with extensive new information and there are additional chapters on characterizing antibodies, antibody engineering, and flow cytometry. As in the original book, the emphasis in this second edition is on providing clear and authoritative protocols with sufficient background information and troubleshooting advice for the novice as well as the experienced investigator.

2013, 847 pp., illus. (32 4C, 103 B&W), appendices, index
Hardcover $260 £160
Paperback $175 £108

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Manipulating the Mouse Embryo
A Laboratory Manual, Fourth Edition

By Richard Behringer, University of Texas, M.D. Anderson Cancer Centre, Marina Gertsenstein, Toronto Centre for Phenogenomics, Transgenic Core and Specialty Resources, Kristina Nagy, Samuel Lunenfeld Research Institute, Mount Sinai Hospital, Toronto, and Andras Nagy, Samuel Lunenfeld Research Institute, Mount Sinai Hospital, Toronto

The fourth edition of the “Mouse Manual”—Manipulating the Mouse Embryo—appears 28 years after the first edition and once again is the definitive reference source on mouse development, transgenesis techniques, and molecular biology. Authors Richard Behringer, Marina Gertsenstein, Kristina Nagy, and Andras Nagy—pre-eminent leaders in their fields—have reorganized and updated this edition to include new information and protocols on:

• assisted reproduction techniques for sperm and embryo cryopreservation
• generation of induced pluripotent stem cells
• isolation, generation, and transplantation of spermatogonial stem cell lines
• in utero electroporation of gene constructs into post-implantation embryos
• vibratome sectioning of live and fixed tissues for imaging thick tissue sections
• whole-mount fluorescent staining methods for three-dimensional visualization.

Techniques regarding recombinant DNA technology and mouse embryonic development from the previous editions have been updated and recast, as has the wealth of information on mouse laboratory strains, mouse housing and breeding, surgical procedures, assisted reproduction, handling of embryos, and micromanipulation setups. The first edition of Manipulating the Mouse Embryo appeared in 1986 as an outgrowth of Cold Spring Harbor Laboratory courses on the molecular embryology of the mouse held in the early 1980s, and authors of the first two editions included Brigid Hogan, Rosa Beddington, Frank Costantini, and Liz Lacy. Mouse embryo manipulation techniques have developed exponentially since the first edition, but then, as now, Manipulating the Mouse Embryo remains the essential practical and theoretical guide for anyone working with mice—students, lab technicians, and investigators.

Due November 2013, 850 pp. (approx.), illus., index
Hardcover $240 £150
Paperback $165 £104

ISBN 978-1-936113-00-2

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Calcium Techniques
A Laboratory Manual

Edited by Jan B. Parys, University of Leuven, Martin Bootman, The Babraham Institute, David I. Yule, University of Rochester, and Geert Bultynck, University of Leuven

Life begins with a surge of calcium ions (Ca\(^{2+}\)) at fertilization, and thereafter, Ca\(^{2+}\) signaling influences nearly every aspect of mammalian development and physiology, from gene expression and cell proliferation to muscle contraction and nerve impulses. To create spatiotemporally distinct Ca\(^{2+}\) signals, cells use a variety of mechanisms to recognize, transport, and buffer Ca\(^{2+}\). Thus, a diverse range of reliable experimental techniques is necessary to study the movement of Ca\(^{2+}\) and the various effectors involved.

This laboratory manual provides step-by-step protocols for studying many facets of Ca\(^{2+}\) signaling, as well as background information on the principles and applications of the techniques. Contributors discuss how to use fluorescent, luminescent, and genetically encoded Ca\(^{2+}\) probes in conjunction with state-of-the-art imaging modalities to characterize Ca\(^{2+}\) signals. Electrophysiological measurements of Ca\(^{2+}\) channel activity are described, as are radioactive Ca\(^{2+}\) flux assays and methods to investigate signaling mediated by specific Ca\(^{2+}\)-mobilizing messengers (IP3, cADPR, and NAADP). Techniques to modulate and suppress intra- and intercellular signals are also provided. Each protocol is complete with a list of required materials, detailed recipes for media and reagents, and troubleshooting advice.

Specific chapters are devoted to Ca\(^{2+}\) signaling techniques in non-mammalian systems, such as plants, yeast, zebrafish, and Xenopus. Methods for assessing Ca\(^{2+}\)-binding kinetics and strategies for developing mathematical models of Ca\(^{2+}\) signaling are also included. Thus, this manual is a comprehensive laboratory resource for biochemists, cell and developmental biologists, and physiologists who are using or looking to expand their repertoire of Ca\(^{2+}\) techniques.

Due November 2013, 500 pp. (approx.), illus., index
Hardcover $150  £86
Paperback $95  £61
ISBN 978-1-621820-78-9

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Cell culture systems for specific neural cell types are essential for studies of their development and function. This laboratory manual provides step-by-step protocols for isolating specific cell populations from rodent tissues and culturing them under conditions that closely resemble those in vivo. The contributors describe in detail how to dissect the brain, spinal cord, and other tissues; how to separate cells using mechanical and enzymatic tissue-dissociation strategies; the use of immunopanning and fluorescence-activated cell sorting (FACS) to enrich the target cell population; and the culture conditions that optimize cell viability and growth. Retinal ganglion cells, motor neurons, dorsal root ganglion cells, astrocytes, oligodendrocytes, and Schwann cells are covered, as are vascular cells such as pericytes and endothelial cells. Myelinating co-cultures of neurons and oligodendrocytes are also described.

The manual includes detailed recipes for media and reagents, tips for avoiding common pitfalls, and advice for designing new immunopanning protocols using tissues from other sources. Many of the protocols are accompanied by freely accessible online movies that demonstrate critical steps of the procedures. This is an essential laboratory companion for all neurobiologists, from the graduate student level upwards.

Due November 2013, 205 pp., illus., index
Hardcover $135 £87
Paperback $89 £57
ISBN 978-1-621820-11-6

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Mouse Models of Cancer
A Laboratory Manual

Edited by Cory Abate-Shen, Herbert Irving Comprehensive Cancer Center, Columbia University College of Physicians and Surgeons, Columbia University Medical Center, Katerina Politi, Yale Cancer Center, Yale University School of Medicine, Lewis Chodosh, Perelman School of Medicine, University of Pennsylvania, and Kenneth P. Olive, Herbert Irving Comprehensive Cancer Center, Columbia University Medical Center

The laboratory mouse is an important model for addressing questions in cancer biology. In recent years, the questions have become more refined, and mouse models are increasingly being used to develop and test cancer therapeutics. Thus, the need for more sophisticated and clinically relevant mouse models has grown, as has the need for innovative tools to analyze and validate them.

This laboratory manual provides cutting-edge methods for generating and characterizing mouse models that accurately recapitulate many features of human cancer. The contributors describe strategies for producing genetic models, including transgenic germline models, gene knockouts and knockins, and conditional and inducible systems, as well as models derived using transposon-based insertional mutagenesis, RNA interference, viral-mediated gene delivery, and chemical carcinogens. Tissue recombination, organ reconstitution, and transplantation methods to develop chimeric, allograft, and xenograft models are covered. Approaches to characterize tumor development, progression, and metastasis in these models using state-of-the-art imaging, histopathological, surgical, and other techniques are also included.

Other chapters cover the use of mouse models to test and optimize drugs in pre-, co-, and post-clinical trials. An appendix specifically addresses the use of mouse cancer models in translational studies and the integration of mouse and human clinical investigations. This manual is therefore an indispensable laboratory resource for all researchers, from the graduate level upwards, who study cancer and its treatment.

Due December 2013, 500 pp. (approx.), illus., index

Hardcover $240 £150
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ISBN 978-1-621820-03-1

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Molecular Cloning: A Laboratory Manual
Fourth Edition

By Michael R. Green, Howard Hughes Medical Institute, University of Massachusetts Medical School and Joseph Sambrook, Peter MacCallum Cancer Institute, Melbourne, Australia

Molecular Cloning: A Laboratory Manual has always been the one indispensable molecular biology laboratory manual for protocols and techniques. The fourth edition of this classic manual preserves the detail and clarity of previous editions as well as the theoretical and historical underpinnings of the techniques presented. Ten original core chapters reflect developments and innovation in standard techniques and introduce new cutting-edge protocols. Twelve entirely new chapters are devoted to the most exciting current research strategies, including epigenetic analysis, RNA interference, genome sequencing, and bioinformatics. This manual is essential for both the inexperienced and the advanced user.

2012, 2,028 pp., illus. (74 4C, 157 2C, and 50 B&W), appendices, index
Cloth (three-volume set) $395 £259
Paperback (three-volume set) $365 £230

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BEST-SELLING MANUALS
RNA molecules participate in and regulate a vast array of cellular processes, and the scientific community is now entering a new era in which some aspect of RNA biology—as a tool, a therapeutic, a diagnostic, or part of a fundamental process—is becoming increasingly important. But initiating RNA research can be intimidating, and without a thorough understanding of the challenges and complexities inherent in handling this fragile nucleic acid, forays into the RNA world can be quite frustrating. *RNA: A Laboratory Manual* provides a broad range of up-to-date techniques so that any investigator can confidently handle RNA and carry out meaningful experiments, from the most basic to the most sophisticated. Originating in four of the field’s most prominent laboratories and written with novices as well as more advanced researchers in mind, this manual provides the necessary background and strategies for approaching any RNA investigation in addition to detailed step-by-step protocols and extensive tips and troubleshooting information. *RNA: A Laboratory Manual* will enable any researcher to approach a wide variety of RNA-related problems with confidence and a high expectation of success.

2011, 586 pp., illus., appendices, index
Hardcover $246 £155
Paperback $169 £107

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Imaging
A Laboratory Manual

Edited by and Series Editor Rafael Yuste, Howard Hughes Medical Institute, Columbia University

In recent years, imaging has rapidly become a tremendously valuable approach in nearly every field of biological research. Finding the right method and optimizing it for data collection can be a daunting process, even for an established imaging laboratory. *Imaging: A Laboratory Manual* is the cornerstone of a new laboratory manual series, designed as an essential guide for investigators who need these visualization techniques. This first volume is meant as a general reference for all fields, and describes the theory and practice of a wide array of imaging methods. From the basic chapters on optics, equipment and labeling to detailed explanations of advanced, cutting-edge methods like PALM, STORM, light sheet and high speed microscopy, *Imaging: A Laboratory Manual* is a vital resource for the modern biology laboratory.

2011, 952 pp., illus., appendices, index
Hardcover $246 £155
Paperback $169 £107
Also available as an Amazon Kindle book.

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Imaging in Neuroscience
A Laboratory Manual

Edited by Fritjof Helmchen, Brain Research Institute, University of Zurich, Switzerland, and Arthur Konnerth, Institute for Neuroscience, Technical University Munich, Germany; Series Editor, Rafael Yuste, Howard Hughes Medical Institute, Columbia University

As imaging technologies have revolutionized research in many areas of biology and medicine, neuroscientists have often pioneered the use of these new visualization techniques. Imaging in Neuroscience: A Laboratory Manual, part of Cold Spring Harbor Laboratory Press’ Imaging series, provides the definitive collection of methods in use in this groundbreaking field. With over 90 chapters, the manual offers a depth of coverage unavailable from any other source. Sections focus on imaging at the molecular level, axons and nerve terminals, spines and dendrites, neurons and circuits in vitro, neurons and circuits in vivo, glia, brain dynamics, and behavior and brain pathology. Protocols range from basic techniques such as maintaining live cells and tissue slices during imaging to recent breakthroughs in optogenetics, uncaging, calcium imaging and imaging neuronal activity. Imaging in Neuroscience: A Laboratory Manual is an essential guide to discovering and implementing these techniques in the neuroscience laboratory.

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The second edition of Live Cell Imaging: A Laboratory Manual expands upon and extends the collection of established and evolving methods for studying dynamic changes in living cells and organisms presented in the well-known first edition. There are 16 new chapters and the 21 updated chapters in this new edition. They include advances in atomic force microscopy, structured illumination microscopy and other 3-D approaches, as well as imaging in single cells in animals and in plants. New analytical options include live high-throughput/high-content screening in mammalian cells and computational analysis of live cell data. The manual presents hands-on techniques as well as background material, and can serve as a text in advanced courses. The first section covers principles and fundamental issues of detection and imaging; the second provides detailed protocols for imaging live systems.

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Cold Spring Harbor Laboratory’s long-running Neurobiology of Drosophila course has trained a generation of neuroscientists, many of whom have become leaders in the field. *Drosophila Neurobiology: A Laboratory Manual* offers the detailed protocols and background material developed by the course instructors to all researchers interested in using *Drosophila* as an experimental model for investigating the nervous system. The manual covers three approaches to the field: Studying Neural Development in *Drosophila melanogaster*, Recording and Imaging in the *Drosophila* Nervous System, and Studying Behavior in *Drosophila*. Techniques described include molecular, genetic, electrophysiological, imaging, behavioral and developmental methods. Written by leading experts from the community, *Drosophila Neurobiology: A Laboratory Manual* is an essential guide for researchers at all levels, from the beginning graduate student through the established primary investigator.

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The mouse has become a standard laboratory model organism, particularly for the study of hematopoiesis, the immune system, and inflammation. Although laboratories studying stem cells, blood, and blood-forming tissues have assimilated many new molecular diagnostic methods, the identification of cell lineages through classical light microscopic techniques is often poorly understood and practiced. *Mouse Hematology* presents a concise review of conventional methods for the preparation, enumeration, and microscopic examination of blood and blood-forming tissues of the laboratory mouse. Along with a short laboratory manual featuring detailed protocols, *Mouse Hematology* includes a DVD of short video demonstrations of the techniques and a poster of blood cell types for easy identification at the microscope. These rapid, inexpensive assessments can save valuable time and resources essential to the design, development, and interpretation of experiments.

2010, 100 pp., illus., appendix, index; poster; DVD
Hardcover $169 £107
Paperback $103 £65
ISBN 978-0-879698-85-0

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**Mouse Peripheral Blood Cells**

A quick reference guide to mouse peripheral blood cells, including brief descriptions of each lineage, total cellularities, and hematological/physiological parameters of interest.
Genetics of Complex Human Diseases
A Laboratory Manual

Edited by Ammar Al-Chalabi, MRC Centre for Neurodegeneration Research, King's College London, and Laura Almasy, Southwest Foundation for Biomedical Research, San Antonio, Texas

Many human diseases—including Alzheimer’s disease, schizophrenia, cancer, and cardiovascular disease—show complex inheritance that requires sophisticated analysis. Genetics of Complex Human Diseases: A Laboratory Manual brings together the tools that geneticists use to find disease genes with the genetic concepts and statistical theories that underpin these research approaches. Topics covered include basic genetics and Mendelian inheritance, statistical methods, genetic epidemiology, linkage studies, transmission disequilibrium test analysis, variance components analysis, genome-wide association studies, copy-number variation, methods for high-throughput genotyping, the complexity of RNA editing, and genetic computer programs. The book’s chapters, written by leading investigators in the field, blend practical information and reviews of each topic, providing both the how and the why of complex disease analysis. Genetics of Complex Human Diseases is an important guide for anyone with an interest in human genetics or who uses genetic techniques in the study of diseases with complex inheritance.

2009, 220 pp., illus., index
Hardcover $162 £102
Paperback $97 £61
ISBN 978-0-879698-82-9
ISBN 978-0-879698-83-6

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Strategies for studying gene regulation mechanisms have changed dramatically over the past several years in light of the emergence of complete genome sequences for many organisms as well as the development of or improvements to technologies such as chromatin immunoprecipitation, RNA interference, microarrays, and proteomics.

The first edition of the highly successful *Transcriptional Regulation in Eukaryotes*, written by Michael Carey and Stephen Smale at UCLA, provided a comprehensive source of strategic, conceptual, and technical information for investigating the complexities of gene regulation at the level of transcription.

With the ever-increasing importance of genome data and the appearance of new and better techniques, the second edition of this book has added a third author, Craig Peterson at the University of Massachusetts Medical School. In addition to a new chapter on the in vitro analysis of chromatin templates for DNA-binding studies and transcription, this second edition has been extensively rewritten and updated to discuss new advances in the field and their impact on gene regulation mechanisms. The second edition retains the approach of the first in covering both the conceptual and practical aspects of how to study the regulation of a newly isolated gene and the biochemistry of a new transcription factor.

*Transcriptional Regulation in Eukaryotes* serves as both a powerful textbook and manual for advanced instruction in molecular biology which

- supplements clearly written text with extensive illustrations
- puts methods in the context of underlying theory
- gives expert recommendations on experimental strategies
- encourages creativity in investigative design
- explains protocols for essential techniques step by step, with extensive advice on troubleshooting
- provides the latest methods in use in the field

This important and unique book is essential reading for anyone pursuing the analysis of gene expression in model systems or disease states, providing underlying theory and perspective to the newcomer and the latest techniques to the expert.

2009, 620 pp., appendix, index

Hardcover $246 £155

Paperback $169 £107

ISBN 978-0-879697-77-8


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*Concepts, Strategies, and Techniques, Second Edition*

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Download copies of the figures from *Transcriptional Regulation in Eukaryotes* at [www.cshteaching.org](http://www.cshteaching.org)
Proteomics: A Cold Spring Harbor Laboratory Course Manual

By Andrew J. Link, Vanderbilt University School of Medicine, Nashville, Tennessee and Joshua LaBaer, Harvard University School of Medicine

Based on a popular course at Cold Spring Harbor Laboratory, this new manual assembles cutting-edge protocols, helpful hints, and lecture notes to teach researchers from a wide variety of disciplines the essential methods of proteomics using state-of-the-art instrumentation. Detailed protocols involving protein microarrays, liquid chromatography, high-throughput cloning of expression constructs, IMAC, mass spectrometry, MALDI–TOF, and MudPIT are provided, along with well-illustrated descriptions of experimental procedures and lists of recommended Web sites and reading material. Proteomics: A Cold Spring Harbor Laboratory Course Manual can be used both as the basis for a course and as a detailed bench manual for those performing indispensable proteomic experiments. It is authored by Andrew J. Link and Joshua LaBaer, both leaders in their fields, who bring complementary expertise to the manual.

2009, 228 pp., illus., appendices, index
Hardcover $154 £97
Paperback $96 £61

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1-855-452-6793  C26
As molecular and cellular biologists move toward nano–techniques for performing experiments on single molecules rather than on populations of molecules, a comprehensive manual on how (and why) to carry out such experiments is needed. *Single-Molecule Techniques: A Laboratory Manual* fills this requirement—it is the first to take researchers who know nothing about single–molecule analyses to the point where they can successfully design and execute appropriate experiments. Geared toward research scientists in structural and molecular biology, biochemistry, and biophysics, the manual will be useful to all who are interested in observing, manipulating, and elucidating the molecular mechanisms and discrete properties of macromolecules. Techniques range from in vivo and in vitro fluorescent–based methods to the use of atomic force microscopy, optical and magnetic tweezers, and nanopores. The book is edited by Paul R. Selvin and Taekjip Ha, two pioneers in the field of experimental biophysics who have made significant contributions to the development and application of single–molecule technologies.

2008, 507 pp., illus., appendix, index
Paperback $169 £107
ISBN 978-0-879697-75-4

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Genetic Variation
A Laboratory Manual


Genetic Variation: A Laboratory Manual is the first compendium of protocols specifically geared towards genetic variation studies, and includes thorough discussions on their applications for human and model organism studies. Intended for graduate students and professional scientists in clinical and research settings, it covers the complete spectrum of genetic variation—from SNPs and microsatellites to more complex DNA alterations, including copy number variation. Written and edited by leading scientists in the field, the early sections of the manual are devoted to study design and generating genotype data, the use of resources such as HapMap and dbSNP, as well as experimental, statistical, and bioinformatic approaches for analyzing the data. The final sections include descriptions of genetic variation in model organisms and discussions of recent insights into human genetic ancestry, forensics, and human variation.

2007, 472 pp., illus., appendix, index
Paperback $165 £104 ISBN 978-0-87969-780-8

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Gene Transfer: Delivery and Expression of DNA and RNA
A Laboratory Manual

Edited by Theodore Friedmann, University of California, San Diego and John Rossi, Beckman Research Institute of the City of Hope, Duarte, California

Understanding gene function and regulation requires rigorous testing in live cells and organisms. Recent advances have provided a variety of new strategies for delivering DNA and RNA into cells and probing their expression, as well as new clinical applications that rely upon the introduction of genetic material. The vast number of available techniques for clinical and laboratory research often makes selecting the optimal method a difficult process. Gene Transfer: Delivery and Expression of DNA and RNA provides the first comprehensive guide to technical approaches for delivering nucleic acids into cells and organisms and of ensuring (even manipulating) appropriate expression. The detailed, step-by-step protocols cover a variety of methods, both well established and newly evolving. These include viral and nonviral methods of gene delivery, transgenic approaches, strategies for the regulation of transgene expression, and modification of the host response. The introductory matter to each chapter includes concise technical and theoretical discussions with considerations for selection of the appropriate system and strategies for delivery.

2007, 793 pp., illus., appendix, index
Paperback $167 £26
Hardcover $263 £42

ISBN 978-087969764-8

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Basic Methods in Microscopy
Protocols and Concepts from Cells
A Laboratory Manual

By David L. Spector, Cold Spring Harbor Laboratory, and Robert D. Goldman, Northwestern University Medical School, Chicago

Imaging has become a vital tool for researchers in all aspects of biology. Recent advances in microscope technology, labeling techniques and gene and protein manipulation methods have led to breakthroughs in our understanding of biological processes. In order to take advantage of these techniques, biologists need to understand the fundamental techniques of microscopy. The methods found here, drawn from the popular laboratory standard manuals Cells: A Laboratory Manual and Live Cell Imaging provide a solid course in the basics of using the microscope in a biology laboratory.

Basic Methods in Microscopy provides an essential guide to light microscopy, fluorescence microscopy, confocal microscopy, multiphoton microscopy and electron microscopy, preparation of tissues and cells, labeling of specimens and analysis of cellular events.

This manual is an important tool for any biology researcher employing imaging as a research method.

2006, 382 pp., illus., index
Paperback $97 £61
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“Methods in Yeast Genetics” is a course that has been offered annually at Cold Spring Harbor Laboratory for the last 30 years. This is an updated edition of the course manual, which provides a set of teaching experiments, along with protocols and recipes for the standard techniques and reagents used in the study of yeast biology. Since the last edition of the manual was published (2000), revolutionary advances in genomics and proteomics technologies have had a significant impact on the field. This updated edition reflects these advances, and also includes new techniques involving vital staining, visualization of green fluorescent protein, new drug resistance markers, high-copy suppression, tandem affinity protein tag protein purification, gene disruption by double-fusion polymerase chain reaction, and many other recent developments.

2005, 230 pp., illus.
Paperback $96 £61
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Drosophila Protocols

By William Sullivan, University of California, Santa Cruz, Michael Ashburner, University of Cambridge, and R. Scott Hawley, University of California, Davis

This exceptional laboratory manual describes thirty-seven procedures most likely to be used in the next decade for molecular, biochemical, and cellular studies on Drosophila. They were selected after extensive consultation with the research community and rigorously edited for clarity, uniformity, and conciseness.

The outstanding features of this protocol collection are:

Scope: The methods included permit investigation of chromosomes, cell biology, molecular biology, genomes, biochemistry, and development.

Depth: Each protocol includes the basic information needed by novices, with sufficient detail to be valuable to experienced investigators.

Format: Each method is carefully introduced and illustrated with figures, tables, illustrations, and examples of the data obtainable.

Added value: The book's appendices include key aspects of Drosophila biology, essential solutions, buffers, and recipes.

An evolution of Michael Ashburner's 1989 classic Drosophila: A Laboratory Manual, this book is an essential addition to the personal library of Drosophila investigators and an incomparable resource for other research groups with goals likely to require fly-based technical approaches.

2000, 728 pp., illus., appendices, index
Paperback $81  £51

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