

METHODS IN MOLECULAR BIOLOGY™

Series Editor
John M. Walker
School of Life Sciences
University of Hertfordshire
Hatfield, Hertfordshire, AL10 9AB, UK

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DNA Recombination

Methods and Protocols

Edited by

Hideo Tsubouchi

*University of Sussex,
Brighton, United Kingdom*

 **Humana Press**

Editor

Hideo Tsubouchi
MRC Genome Damage and Stability Centre
University of Sussex
Science Park Road, Falmer
Brighton, BN1 9RQ
United Kingdom
h.tsubouchi@sussex.ac.uk

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Preface

Homologous recombination has been intensively studied in budding yeast. I think we are extremely lucky to find that homologous recombination is exceptionally robust in this organism, making it an ideal model to study this process. Historically, the availability of powerful genetics in this simple, unicellular organism has enabled the isolation of genes that play key roles in homologous recombination, and we have learnt a lot about homologous recombination using this organism. Homologous recombination is important in various aspects of DNA metabolism, including damage repair, replication, telomere maintenance, and meiosis. We also now know that key players in homologous recombination identified and characterized in yeast, such as proteins encoded by the genes belonging to the so-called RAD52 group, are well conserved among eukaryotic species, including humans. This offers promise that further in-depth characterization of homologous recombination using yeast will help provide the basic framework for understanding the universal mechanism(s) of homologous recombination conserved in eukaryotes. When asked to edit a book about methods for studying homologous recombination, I decided to include chapters that cover recent techniques that best utilize the advantages of the yeast system, with the belief that yeast will keep serving as a great model organism to study homologous recombination.

On the other hand, there is a group of genes involved in recombination that are apparently found only in higher eukaryotes, such as BRCA2, indicating the presence of an extra layer of mechanistic complexity in these organisms. Obviously, the most straightforward approach to study these mechanisms is to use models in which these particular mechanisms exist. From this point of view, chapters for studying recombination using higher eukaryotes have also been included.

Although we have gained significant understanding of the entity underlying homologous recombination, I have to say that we still do not know much about it when we see it as a “micro machine” that is incredibly efficient at finding similarity between two DNA molecules inside a cell. Obviously, a necessary step in the direction of understanding this process is to isolate the machine and let it work in a test tube. Understanding the design by studying the appearance and behavior of the machinery as a single molecule will be an important milestone toward understanding the mechanism of action of the machinery. Almost as important is to learn how the machinery behaves inside living cells. In recent years, this approach has flourished due to advances in microscopy and the availability of various fluorescent proteins. Techniques covering these topics have been included.

Yeast genetics has successfully provided a framework for the mechanism of homologous recombination. Now the question is, what can we do next to bring it to the next level of understanding? This is a question I ask myself, but I believe it is more or less a question for anyone who is enthusiastic about understanding this very fascinating phenomenon. I hope this protocol book will prove useful for this purpose. Finally, I would like to thank all the contributors who willingly agreed to share their expertise/knowledge. Needless to say, this book would not exist without their effort.

Hideo Tsubouchi

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Contributors

- CAROLINE ADELFAK • *Max-Planck-Institute for Molecular Genetics, Berlin, Germany*
- ANDRÉS AGUILERA • *Centro Andaluz de Biología Molecular y Medicina Regenerativa, Universidad de Sevilla-CSIC, Sevilla, Spain*
- JASVINDER S. AHUJA • *Department of Biological, Geological and Environmental Sciences, Center for Gene Regulation in Health and Disease, Cleveland State University, Cleveland, OH, USA*
- VERA BATRAK • *Independent Scientist, Istra, Moscow Region, Russia*
- GRACE C. BAZAN • *Biological Sciences, California Polytechnic State University, San Luis Obispo, CA, USA*
- ARTEM BLAGODATSKI • *Institute of Protein Research, Russian Academy of Sciences, Russian Federation, Moscow, Russia*
- HANNAH G. BLITZBLAU • *Whitehead Institute for Biomedical Research, Cambridge, MA, USA*
- G. VALENTIN BÖRNER • *Department of Biological, Geological and Environmental Sciences, Center for Gene Regulation in Health and Disease, Cleveland State University, Cleveland, OH, USA*
- ERIKA BRUNET • *Muséum National d'Histoire Naturelle, Paris, France*
- JEAN-MARIE BUERSTEDDE • *Independent Scientist, Hildesheim, Germany*
- DMITRY V. BUGREEV • *Department of Biochemistry and Molecular Biology, Drexel University College of Medicine, Philadelphia, PA, USA*
- KERSTIN BYSTRICKY • *Laboratoire de Biologie Moléculaire Eucaryote (LBME), Université de Toulouse, Toulouse, France*
- FRANCESCA CAVALLO • *Department of Public Health and Cell Biology, Section of Anatomy, University of Rome Tor Vergata, Rome, Italy*
- STACY Y. CHEN • *Department of Obstetrics, Gynecology, and Reproductive Sciences, University of California, San Francisco, CA, USA*
- FRANCESCA COLE • *Developmental Biology Program, Memorial Sloan-Kettering Cancer Center, New York, NY, USA*
- VINCENZO COSTANZO • *Clare Hall Laboratories, London Research Institute, Hertfordshire, UK*
- JAN DROUAUD • *Institut Jean-Pierre Bourgin, UMR1318 INRA-AgroParisTech, Versailles Cedex, France; Institut National de Recherche, Agronomique, Centre de Versailles-Grignon Route de St-Cyr (RD10), Versailles Cedex, France*
- NADINE ECKERT-BOULET • *Department of Biology, University of Copenhagen, Copenhagen, Denmark*
- KIRK T. EHMTSEN • *Department of Microbiology, University of California, Davis, CA, USA*
- ANASTASIYA EPSTEIN • *Department of Biochemistry, New York University School of Medicine, New York, NY, USA*
- SARAH FARMER • *MRC Genome Damage and Stability Centre, University of Sussex, Sussex, UK*

- ILYA J. FINKELSTEIN • *Department of Biochemistry and Molecular Biophysics, Columbia University, New York, NY, USA*
- JENNIFER C. FUNG • *Department of Obstetrics, Gynecology, and Reproductive Sciences, University of California, San Francisco, CA, USA*
- BELÉN GÓMEZ-GONZÁLEZ • *Centro Andaluz de Biología Molecular y Medicina Regenerativa, Universidad de Sevilla-CSIC, Sevilla, Spain*
- ERIC C. GREENE • *Department of Biochemistry and Molecular Biophysics, Columbia University, New York, NY; Howard Hughes Medical Institute, Chevy Chase, MD, USA*
- JILL M. GRIMME • *US Army Engineer Research Development Center, Construction Engineering Research Laboratory, Champaign, IL, USA*
- EDGAR HARTSUIKER • *North West Cancer Research Fund Institute, Bangor University, Bangor, UK*
- YOSHITAMI HASHIMOTO • *Clare Hall Laboratories, London Research Institute, Hertfordshire, UK*
- WOLF-DIETRICH HEYER • *Department of Microbiology and Department of Molecular and Cellular Biology, University of California, Davis, CA, USA*
- KENNETH J. HILLERS • *Biological Sciences, California Polytechnic State University, San Luis Obispo, CA, USA*
- SEIKI HIRANO • *Weatherall Institute of Molecular Medicine, University of Oxford, Oxford, UK*
- ANDREAS HOCHWAGEN • *Whitehead Institute for Biomedical Research, Cambridge, MA, USA*
- NANCY M. HOLLINGSWORTH • *Department of Biochemistry and Cell Biology, Stony Brook University, New York, NY, USA*
- SCOTT HOUGHTALING • *Department of Molecular Genetics and Microbiology, University of New Mexico School of Medicine, Albuquerque, NM, USA*
- HIROSHI IWASAKI • *School and Graduate School of Bioscience and Biotechnology, Tokyo Institute of Technology, Tokyo, Japan*
- MARIA JASIN • *Developmental Biology Program, Memorial Sloan-Kettering Cancer Center, New York, NY, USA*
- HIROYUKI KITAO • *Department of Molecular Oncology, Kyushu University, Kyushu, Japan*
- HANNAH L. KLEIN • *Department of Biochemistry, New York University School of Medicine, New York, NY, USA*
- YOUNGHO KWON • *Department of Molecular Biophysics and Biochemistry, Yale University School of Medicine, New Haven, CT, USA*
- IMEN LASSADI • *Laboratoire de Biologie Moléculaire Eucaryote, Université de Toulouse, Toulouse, France*
- WING-KIT LEUNG • *MRC Genome Damage and Stability Centre, University of Sussex, Sussex, UK*
- MICHAEL LISBY • *Department of Biology, University of Copenhagen, Copenhagen, Denmark*
- JIE LIU • *Department of Microbiology, University of California, Davis, CA, USA*
- HSIAO-CHI LO • *Department of Biochemistry and Cell Biology, Stony Brook University, New York, NY, USA*
- WENJIAN MA • *Chromosome Stability Section, National Institute of Environmental Health Sciences (NIEHS), NIH, Research Triangle Park, NC, USA*

- ANNA MALKOVA • *Biology Department, Indiana University Purdue University, Indianapolis, IN, USA*
- ALEXANDER V. MAZIN • *Department of Biochemistry and Molecular Biology, Drexel University College of Medicine, Philadelphia, PA, USA*
- OLGA M. MAZINA • *Department of Biochemistry and Molecular Biology, Drexel University College of Medicine, Philadelphia, PA, USA*
- CHRISTINE MÉZARD • *Institut Jean-Pierre Bourgin, Versailles Cedex, France*
- KUNTAL MUKHERJEE • *School of Biology, Georgia Institute of Technology, Atlanta, GA, USA*
- YASUTO MURAYAMA • *Cancer Research UK, London Research Institute, London, UK*
- KENTARO NABESHIMA • *Department of Cell and Developmental Biology, University of Michigan, Medical School, Ann Arbor, MI, USA*
- WATARU NAKAI • *Chromosome Stability Section, National Institute of Environmental Health Sciences (NIEHS), NIH, Research Triangle Park, NC, USA*
- KOJI NAKANISHI • *Developmental Biology Program, Memorial Sloan-Kettering Cancer Center, New York, NY, USA*
- MARY ANN OSLEY • *Department of Molecular Genetics and Microbiology, University of New Mexico School of Medicine, Albuquerque, NM, USA*
- MICHAEL A. RESNICK • *Chromosome Stability Section, National Institute of Environmental Health Sciences (NIEHS), NIH, Research Triangle Park, NC, USA*
- MATTHEW J. ROSSI • *Department of Biochemistry and Molecular Biology, Drexel University College of Medicine, Philadelphia, PA, USA*
- RODNEY ROTHSTEIN • *Department of Genetics and Development, Columbia University Medical Center, New York, NY, USA*
- JOSÉ F. RUIZ • *Centro Andaluz de Biología Molecular y Medicina Regenerativa, Universidad de Sevilla-CSIC, Sevilla, Spain*
- HARRY SCHERTHAN • *Bundeswehr Institute of Radiobiology, affiliated to the University of Ulm, Munich, Germany; Max-Planck-Institute for Molecular Genetics, Berlin, Germany*
- HILARIE A. SEHORN • *Department of Genetics and Biochemistry, Clemson University, Clemson, SC, USA*
- MICHAEL G. SEHORN • *Department of Genetics and Biochemistry, Clemson University, Clemson, SC, USA*
- YING SHEN • *School of Biology, Georgia Institute of Technology, Atlanta, GA, USA*
- JESSICA SNEEDEN • *Department of Microbiology, University of California, Davis, CA, USA*
- MARIA SPIES • *Department of Biochemistry, Howard Hughes Medical Institute, University of Illinois, Urbana-Champaign, Urbana, IL, USA*
- FRANCESCA STORICI • *School of Biology, Georgia Institute of Technology, Atlanta, GA, USA*
- SAMANTHA STUCKEY • *School of Biology, Georgia Institute of Technology, Atlanta, GA, USA*
- PATRICK SUNG • *Department of Molecular Biophysics and Biochemistry, Yale University School of Medicine, New Haven, CT, USA*
- MINORU TAKATA • *Laboratory of DNA Damage Signaling, Department of Late Effects Studies, Kyoto University, Kyoto, Japan*
- HIDEO TSUBOUCHI • *MRC Genome Damage and Stability Centre, University of Sussex, Brighton, UK*

TOYOKO TSUKUDA • *Department of Molecular Genetics and Microbiology, University of New Mexico School of Medicine, Albuquerque, NM, USA*

JIM WESTMORELAND • *Chromosome Stability Section, National Institute of Environmental Health Sciences (NIEHS), NIH, Research Triangle Park, NC, USA*

WILLIAM D. WRIGHT • *Department of Microbiology, University of California, Davis, CA, USA*

XIAO-PING ZHANG • *Department of Microbiology, University of California, Davis, CA, USA*

WEIXING ZHAO • *Department of Molecular Biophysics and Biochemistry, Yale University School of Medicine, New Haven, CT, USA*

XIUZHONG ZHENG • *Department of Biochemistry, New York University School of Medicine, New York, NY, USA*