Methods in Enzymology

Volume 152

Guide to Molecular Cloning Techniques

EDITED BY

Shelby L. Berger

Alan R. Kimmel

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Preface

With the birth of recombinant DNA technology, the fields of nucleic acid biochemistry and molecular biology entered an era marked by dramatic and innovative changes in methodology, rapid growth, and an altered perception of how living systems could be studied. As additional possibilities for applying these techniques were realized, it became evident that virtually every area of the life sciences would benefit. For the specialist, this was a very exciting period, but for those who wished to solve specific problems without first becoming full-time molecular biologists, the explosive increase in knowledge was overwhelming. Clearly, the new technology had to be harnessed to serve biologists regardless of scientific background.

This volume represents our contribution. Our aim is to meet the needs of investigators entering molecular biology from other fields and to orient students joining this discipline for the first time. We envisaged a self-contained, concise compendium of state-of-the-art methods that might also appeal to experienced individuals. To impose order on this complex body of information, the book progresses from the basic techniques underlying much of recombinant DNA technology to a series of sections, each addressing a commonly met problem. Topics include genomic cloning, preparation and characterization of mRNA, cDNA cloning, screening libraries, and confirming the identity of selected clones.

The *Guide* contains reliable methods written by leaders in the field. Many articles contrast different approaches for accomplishing the same task in order to highlight strengths and weaknesses in a side-by-side comparison. Others present only the method that was deemed superior. To assist the user, we have been assured that recommended vectors and strains that are not commercially available will be provided by the authors.

Because molecular cloning requires precise attention to detail, the book is heavily edited in the form of cross referencing, Editors' Notes, the Process Guide, and overviews. Pains have been taken to allow the reader to pick and choose among articles without loss of continuity. Cross-referencing helps to clarify the relationships among articles. So, too, do the Editors' Notes. The Process Guide is another integrative device in which fundamental processes in molecular biology are indexed for ready accessibility. Since some methods are used frequently to achieve not quite identical ends, the reader can locate at a glance those that should be considered before choosing a specific technique. Finally,

the volume contains overviews to five of the major sections to introduce concepts and strategies and to aid in rapid recognition of relevant material for a particular task.

Within the framework of a one-volume format, choices had to be made. For example, *Drosophila* and *Saccharomyces*, organisms for which specialized methods are abundant, have been short changed. Separate volumes devoted exclusively to these subjects are required for adequate coverage. Mutagenesis has not been emphasized but is covered elsewhere in this series. Some attractive methods were not included because their utility and reproducibility are not firmly established. The *Guide* is primarily intended as an efficient means toward obtaining and characterizing a clone. Within this narrow scope, the contents were chosen based on what would be most important for most investigators most of the time.

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Process Guide

This listing contains the location by chapter number of specific methods for which protocols have been provided. Generally, methods that are mentioned but not presented in detail, or components of a reaction, are not listed here. Thus, Filling in, Nick translation, and Primer extension are in the Process Guide but DNA polymerase I and Klenow fragment require consultation of the Subject Index.

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