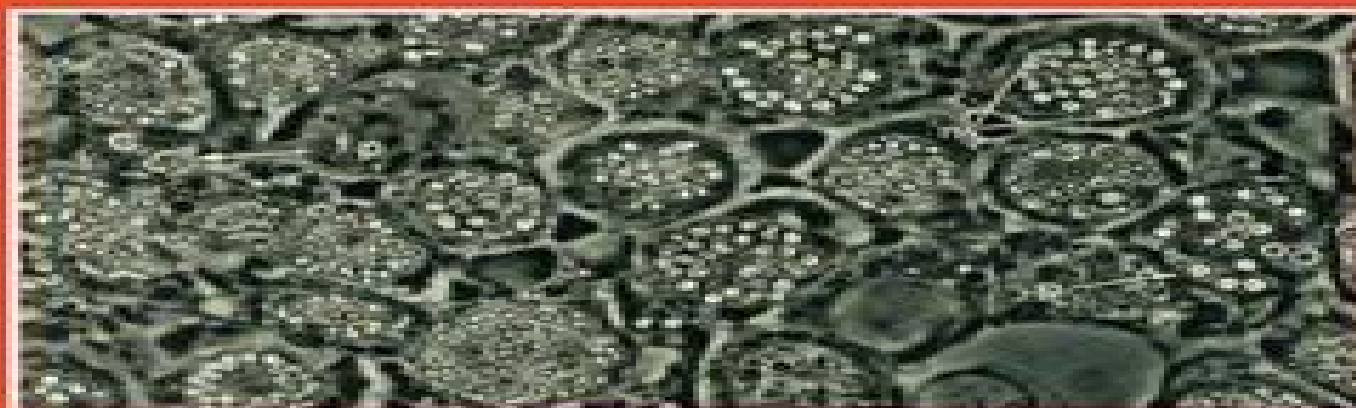


Methods in Molecular Biology

Volume 39

BACULOVIRUS EXPRESSION PROTOCOLS

Edited by
Christopher D. Richardson



Baculovirus Expression Protocols

Linda King



Baculovirus Expression Protocols:

Baculovirus Expression Protocols Christopher D. Richardson, 2008-02-02 Baculovirus Expression Protocols offers both industrial and university based researchers a comprehensive compilation of the latest baculovirus techniques along with step by step instructions and time saving techniques The contributors leading authorities in the field present the assorted expression plasmids currently in use guide the reader through the process of generating and selecting recombinant virus and describe specific examples of recombinant protein production and purification The emphasis is on alternative and simpler screening techniques for the selection of recombinant baculovirus The book also surveys the various insect cell lines currently compatible with the baculovirus system Highlights include production of recombinant virus using linearized DNA and vectors that contain a b galactosidase indicator and a complete list of expression vectors currently available Detailed descriptions for the scale up of protein production using spinner flasks bioreactors and insect larvae are also included Baculovirus Expression Protocols offers both industrial and university based researchers an outstanding collection of reproducible step by step laboratory protocols It will immediately become indispensable for anyone working with baculoviruses and their application in the expression of recombinant proteins in insect cells

Baculovirus and Insect Cell Expression Protocols David W. Murhammer, 2010-11-19 Baculovirus Expression Protocols Second Edition provides the detailed steps required to perform the techniques involved with the use of baculoviruses and insect cell culture and discusses problems that may be encountered This newly revised and updated edition provides a cutting edge step by step guide for biochemists molecular biologists biochemical engineers and others using the BEVS and or insect cells for producing recombinant proteins Furthermore Baculovirus Expression Protocols Second Edition provides assistance to scientists and engineers interested in developing and producing baculovirus insecticides Authors list all available insect cell lines and provide methods for isolating new cell lines The procedures involved in producing products both at laboratory scale and large scale are discussed as well as the production in insect larvae It is hoped that this volume will not only aid the user in successfully completing the tasks described herein but will also stimulate the development of improved techniques and new applications of baculoviruses and insect cell culture Timely and thorough this volume will become an essential reference for anyone using baculoviruses and insect cell culture

Baculovirus and Insect Cell Expression Protocols. Methods in Molecular Biology David W. Murhammer, 2007 Baculovirus Expression Protocols is a detailed guide for using the baculovirus expression vector system BEVS and or insect cells to produce recombinant proteins Procedures for the development and production of baculovirus insecticides at both laboratory and large scale are described in the kind of step by step format that the Methods in Molecular Biology series helped standardize This book is a one stop source for information on baculoviruses and includes sections on using modified baculoviruses to express genes in mammalian cells using *Drosophila* cell lines isolating new cell lines and developing serum free medium Specific applications including using

baculovirus and insect cell system to study apoptosis are also discussed This updated and expanded edition of Baculovirus Expression Protocols provides a perfect lab manual for biochemists molecular biologists biochemical engineers and others for using the BEVS or insect cell lines to produce recombinant proteins *Baculovirus* Manon M.J. Cox,2024-07-01 This detailed volume provides up to date guidance on techniques used to work with baculoviruses and insect cells The book provides basic methods to create recombinant baculoviruses to improve productivity to produce a variety of products to purify products to quantify baculovirus stocks or to quantify product produced and it concludes with alternative uses of either baculovirus or insect cells as tools Written for the highly successful Methods in Molecular Biology series chapters include introductions to their respective topics lists of the necessary materials and reagents step by step and readily reproducible laboratory protocols and tips on troubleshooting and avoiding known pitfalls Authoritative and comprehensive Baculovirus Methods and Protocols serves as an ideal guide for researchers looking to overcome some of the limitations associated with the early baculoviral vectors and cell lines Gene Transfer and Expression Protocols Edward J. Murray,2008-02-02 Biology is the study of living things The classical approach might be described as holistic and descriptive whereas the modern molecular proach aims to be investigative reductionist and mechanistic Genes contain all the information for the structure of all living things thus the understanding of how genes are regulated is an important step toward understanding the nature of living things The study of gene regulation has been made more tractable by the design of simple expe mental models in which a single gene can be isolated from the milieu of the organism The new science of molecular biology has introduced techniques that permit the design of such experimental models In sence the genome of the organism is dissected in such a manner that specific genes may now be introduced into an appropriate cell line Subsequent analysis of the proteins expressed from the genes under study results in the identification of the regulatory DNA sequences

Methods in Molecular Biology: Baculovirus expression protocols John M. Walker,1984 **The Baculovirus Expression System** Linda King,2012-12-06 The decision to write a book about the practical aspects of the baculovirus expression system stems from the numerous phone calls for help we have had and from the many visitors to our labora tories requiring assistance to find the elusive polyhedrin negative virus containing their favourite gene We have also organized two expression system workshops and from the manuals we wrote for these it seemed a logical progression to extend them into book form We appreciate that those who are old hands at the baculovirus expression system may have differing views on some of our procedures but the methods in this book are presented in the light of our own experiences in the laboratory and from our practical workshops and we hope that the book will be especially useful to those new to the system The first three chapters give the background information to the baculovirus expression system and includes advice on how to choose the right transfer vector and discusses the various methods that are available to select recombinant viruses The practical chapters concentrate on those aspects which are novel to the baculovirus system insect cell culture virus amplification and

titration etc and in general leave the standard molecular biological techniques to the other excellent laboratory manuals that are available. However for completeness sake and to avoid constant reference to other manuals we have included brief details of some standard techniques where they are integral to the success of the baculovirus protocols.

Glycoviropology Protocols Richard J. Sugrue, 2008-01-18 Glycoviropology Protocols reviews the increasing importance of glycosylation to the field of viropology as well as virus replication. The chapters provide an overview of glycosylation in relation to virus infection and the generic techniques that are used to analyze and characterize glycoproteins.

Baculovirus Expression Vectors David R. O'Reilly, Lois K. Miller, Verne A. Luckow, 1994 Baculoviruses have proven to be the most powerful and versatile eukaryotic expression vectors available. This unique laboratory manual is designed to help both beginning and experienced researchers construct and use baculovirus vector systems. It simplifies selection of the most appropriate baculovirus vector design for a given problem then describes each step of the implementation process from vector construction to large scale protein production. The book provides an understanding of how the vectors work a biological overview of cells viruses plasmids and promoters guidelines for choosing optimum vectors protocols for growing insect cells and recombinant viruses methods of analyzing protein products and scaling up protein production techniques for producing proteins in insect larvae and easy to use maps charting available expression vectors. This comprehensive approach has many benefits for researchers and students alike. It allows them to understand how and why the vector system works and offers a rapid comparison of options for choosing the right virus plasmid or promoter for vector design and construction with a minimum amount of lost time. The manual is an invaluable resource for every individual engaged in the production of proteins for any purpose.

Antibody Engineering Protocols Sudhir Paul, 2008-02-02 This comprehensive collection of recently developed methods for producing new antibody reagents by immunization and recombinant DNA techniques contains ready to use protocols that illuminate current areas of research on antibody structure functions and applications. The methods can be applied in basic immunological studies involving antibody specificity catalysis and evolution and in the isolation of rare antibodies by phage display technology and the engineering of new antibodies by mutagenesis. They offer insight into new ways of developing clinically useful antibody reagents. Antibody Engineering Protocols constitutes a single source volume for laboratory investigators who want to minimize extensive literature and methodology searches and to work productively in their fields with reproducible step by step protocols.

NMDA Receptor Protocols Min Li, 2008-02-03 Min Li and a panel of hands on experimentalists detail state of the art molecular techniques for studying NMDA ligand gated ion channels and developing assays for nontherapeutic lead selection. The topics range from cDNA cloning to in vitro and in vivo investigation of the channel complex in the mammalian brain. Additional topics include the biochemical analysis of the channel protein and the construction of various heterologous systems for both basic research and high throughput screens HTS for pharmaceutical chemicals. Although the focus is on NMDA receptors the methods are applicable to other ligand gated ion channels and with

some modification may be extended to related membrane signaling receptors NMDA Receptor Protocols offers today's scientists powerful methods for basic research on NMDA receptor structure and function as well as enormous opportunities for clinical investigation toward the development of novel bioactive compounds

Cytochrome P450 Protocols Ian R. Phillips, Elizabeth A. Shephard, 2008-02-02 In *Cytochrome P450 Protocols* Ian Phillips and Elizabeth Shephard assemble a comprehensive collection of cutting edge techniques for the investigation of cytochromes P450. Described in detail by hands on experimentalists for easy reproducibility these methods include spectral analysis purification and enzymatic assays expression in heterologous systems and the production and use of antibodies as well as methods for quantification of gene expression transfection of hepatocytes and for the investigation of DNA protein interactions and genetic polymorphisms. In addition because of the growing importance of in vitro systems in pharmacological toxicology the book contains techniques for the culture of rodent and human hepatocytes and human epidermis. Cytochrome P450 induction as a biomarker for environmental pollution and the generation of mice with targeted gene disruptions complete this exhaustive collection of core techniques. *Cytochrome P450 Protocols* includes in one volume both state of the art and classic methods that have not been superseded but remain extremely useful. The collection provides both novice and experienced researchers across many fields toxicology pharmacology environmental biology biochemistry and molecular biology all the tools needed to elucidate the crucial biological role played by cytochromes P450 in the metabolism of therapeutic drugs chemical carcinogens and environmental pollutants

PCR Cloning Protocols Bing-Yuan Chen, Harry W. Janes, 2008-02-05 *PCR Cloning Protocols* Second Edition updates and expands Bruce White's best selling *PCR Cloning Protocols* 1997 with the newest procedures for DNA cloning and mutagenesis. Here the researcher will find readily reproducible methods for all the major aspects of PCR use including PCR optimization computer programs for PCR primer design and analysis and novel variations for cloning genes of special characteristics or origin with emphasis on long distance PCR and GC rich template amplification. Also included are both conventional and novel enzyme free and restriction site free procedures to clone PCR products into a range of vectors as well as state of the art protocols to facilitate DNA mutagenesis and recombination and to clone the challenging uncharacterized DNA flanking a known DNA fragment

Gene Isolation and Mapping Protocols Jacqueline Boulton, 2008-02-02 An unprecedented collection of all the most up to date techniques for gene isolation and mapping including the latest methods for gene characterization using database analyses. This collection of thoroughly tested recipes also includes chapters for the computational analysis of novel cDNA sequences with up to the minute information on basic sequence analysis sequence similarity searches exon detection and similarity searches and the prediction of gene function. Its state of the art methods constitute indispensable tools for all scientists engaged in the search for specific disease genes or in the general advancement of the human genome project

Basic Cell Culture Protocols Jeffrey W. Pollard, John M. Walker, 1997 Now completely revised and updated from the original much acclaimed and bestselling first edition *Basic Cell*

Culture Protocols 2nd ed offers today's most comprehensive collection of easy to follow cutting edge protocols for the culture of a wide range of animal cells. Its authoritative contributors provide explicit step by step instructions along with extensive notes and tips that allow both experts and beginners to successfully achieve their desired results. Topics range from basic culture methodology to strategies for culturing previously uncultured cell types and hard to culture differentiated cells. Methods are also provided for the analysis of living cells by FACS, video microscopy and confocal microscopy. Like the first edition, this book should be in every cell culture laboratory and be of use to all who use cell cultures in research. **Plant**

Cell Electroporation And Electrofusion Protocols Jac A. Nickoloff, 2008-02-02. Gene transfer is an essential technology for improving our understanding of gene structure and function. Although there are many methods by which DNA may be introduced into cells including heat and chemical treatments and microinjection, electroporation has been found to be the most versatile gene transfer technique. Electroporation is effective with a wide variety of cell types including those that are difficult to transform by other means. For many cell types, electroporation is either the most efficient or the only means known to effect gene transfer. The early and broad success of electric field mediated DNA transfer soon prompted researchers to investigate electroporation for transferring other types of molecules into cells including RNA, enzymes, antibodies and analytic dyes. The first section of **Plant Cell Electroporation and Electrofusion Protocols** includes two chapters that serve as a guide to theoretical and practical aspects of electroporation and will be of particular interest to those developing protocols for as yet untested species or cell types and a third chapter that describes commercially available electroporation instruments. The remaining chapters describe well tested protocols for DNA electrotransfection, electroporation of other biomolecules or cell electrofusion. These chapters also include brief discussions of alternatives to electric field based methods, citing the advantages and limitations of the various methods for achieving specific goals. **Electroporation**

Protocols for Microorganisms Jac A. Nickoloff, 2008-02-02. Electroporation is one of the most widespread techniques used in modern molecular genetics. It is most commonly used to introduce DNA into cells for investigations of gene structure and function and in this regard, electroporation is both highly versatile, being effective with nearly all species and cell types and highly efficient. For many cell types, electroporation is either the most efficient or the only means known to effect gene transfer. However, exposure of cells to brief high intensity electric fields has found broad application in other aspects of biological research and is now routinely used to introduce other types of biological and analytic molecules into cells to induce cell-cell fusion and to transfer DNA directly between different species. The first seven chapters of **Electroporation Protocols for Microorganisms** describe the underlying theory of electroporation, the commercially available instrumentation and a number of specialized electroporation applications such as cDNA library construction and interspecies DNA electrotransfer. Each of the remaining chapters presents a well developed method for electrotransformation of a particular bacterial, fungal or protist species. These chapters also serve to introduce those new to the field the important research questions that are

currently being addressed with particular organisms highlighting both the major advantages and limitations of each species as a model organism and explaining the roles that electroporation has played in the development of the molecular genetic systems currently in use *The Baculoviruses* Lois K. Miller, 2013-06-29 The past decade has witnessed an explosion of information on the molecular biology of insect viruses and a frenzy of activity in applying this information to medicine and agriculture Genetically engineered baculoviruses are presently being tested for commercial use as pesticides and the study of such viruses is also revealing remarkable insights into basic cellular processes such as apoptosis This comprehensive volume provides readers with knowledge of basic and applied baculovirology so that current literature in the field can be appreciated

Protein Kinase C Protocols Alexandra C. Newton, 2008-02-03 Since the discovery that protein kinase C PKC transduces the abundance of signals that result in phospholipid hydrolysis this enzyme has been at the forefront of research in signal transduction Protein Kinase C Protocols covers fundamental methods for studying the structure function regulation subcellular localization and macromolecular interactions of PKC Protein Kinase C Protocols is divided into 11 sections representing the major aspects of PKC regulation and function Part I contains an introduction and a historical perspective on the discovery of PKC by Drs Yasutomi Nishizuka and Ushio Kikkawa Part II describes methods to purify PKC Part III describes the standard methods for measuring PKC activity its enzymatic activity and its stimulus dependent translocation from the cytosol to the membrane Part IV describes methods for measuring the membrane interaction of PKC in vivo and in vitro Part V provides methodologies and techniques for measuring the phosphorylation state of PKC including a protocol for measuring the activity of PKC's upstream kinase PDK 1 Novel methods for identifying substrates are described in Part VI Part VII presents protocols for expressing and analyzing the membrane targeting domains of PKC Part VIII provides a comprehensive compilation of methods used to identify binding partners for PKC Part IX describes pharmacological probes used to study PKC The book ends with a presentation of genetic approaches to study PKC Part X and a discussion of approaches used to study PKC in disease Part XI **Animal Cell Electroporation and Electrofusion Protocols** Jac A. Nickoloff, 2008-02-03 The ability to introduce macromolecules into animal cells including DNA RNA proteins and other bioactive compounds has facilitated a broad range of biological studies from biochemistry and biophysics to molecular biology cell biology and whole animal studies Gene transfer technology in particular will continue to play an essential role in studies aimed at improving our understanding of the relationships between the gene structure and function and it has important practical applications in both biotechnology and biomedicine as evidenced by the current intense interest in gene therapy Although DNA and other macromolecules may be introduced into cells by a variety of methods including chemical treatments and microinjection electroporation has proven to be simpler to perform more efficient and effective with a wider variety of cell types than other techniques The early and broad success of electric field mediated DNA transfer soon prompted researchers to investigate electroporation for transferring other types of molecules into cells including RNA

enzymes antibodies and analytic dyes Animal Cell Electroporation and Electrofusion Protocols begins with three chapters that describe the theoretical and practical aspects of electroporation including a review of the commercially available instrumentation These introductory chapters will be of particular interest to those new to electric field technologies and to those developing protocols for as yet untested species or cell types Nineteen chapters follow that present well tested protocols for electroporation of proteins and DNA into insect fish and mammalian cells

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