

Methods in Molecular Biology™

VOLUME 116

Protein Lipidation Protocols

Edited by
Michael H. Gelb



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Protein Lipidation Protocols

Christine Schneider



Protein Lipidation Protocols:

Protein Lipidation Protocols Michael H. Gelb, 2008-02-03 In *Protein Lipidation Protocols* Michael Gelb brings together a collection of readily reproducible techniques for studying protein lipidation the covalent attachment of lipids to proteins These cutting edge methods many never published before in a hands on format deal with glycosyl phosphatidylinositol GPI containing compounds protein fatty acylation and protein prenylation Included are novel techniques for determining the chemical structure of GPI anchors for radiolabeling the prenyl groups of protein in eukaryotic cells a tool for developing inhibitors of the protein farnesyltransferase and for an exciting lysosomal enzyme that cleaves fatty acyl groups from proteins the first fatty acylase discovered *Protein Lipidation Protocols* offers biochemists cell and molecular biologists medicinal chemists and pharmaceutical researchers state of the art tools for understanding the complex biochemistry of protein lipidation as well as catalyzing the development of many important new biopharmaceuticals including anticancer drugs

Protein Lipidation Maurine E. Linder, 2019 This volume explores techniques used to detect lipids attached to proteins to analyze the function of lipid modifications and to characterize the enzymes that add and remove lipids from proteins The book is organized into seven parts Part One describes chemically based strategies to identify substrates for protein lipidation that can be applied to individual proteins or globally using proteomics Part Two focuses on the enzymes that remove fatty acids from proteins and provides methods to monitor protein biogenesis and palmitate turnover Part Three addresses biochemical and cellular characterization of DHHC S acyltransferases a family of enzymes with 23 members encoded by the human genome Part Four presents the SwissPalm 2 database and tips on how to use it effectively Part Five focuses on fatty acylation that occurs in the lumen of the secretory pathway Parts Six and Seven conclude the book with methods to produce and assay lipid modified and integral membrane proteins Written in the highly successful *Methods in Molecular Biology* series format chapters include introductions to their respective topics lists of the necessary materials and reagents step by step readily reproducible laboratory protocols and tips on troubleshooting and avoiding known pitfalls Cutting edge and authoritative *Protein Lipidation Methods and Protocols* is a valuable resource for experts in the field and for investigators who encounter protein lipidation through their research on a particular cellular process or favorite protein **Eicosanoid**

Protocols Elias A Lianos, 2008-02-03 **Immunocytochemical Methods and Protocols** Lorette C. Javois, 2008-02-02 Lorette Javois timely new 2nd edition revises and updates her widely acclaimed collection of step by step immunocytochemical methods one that is now used in many biological and biomedical research programs The methods are designed for researchers and clinicians who wish to visualize molecules in plant or animal embryos tissue sections cells or organelles In addition to cutting edge protocols for purifying and preparing antibodies light microscopic analysis confocal microscopy FACS and electron microscopy this revised edition contains many new methods for applying immunocytochemical techniques in the clinical laboratory and in combination with in situ hybridization Chaperonin Protocols Christine

Schneider,2008-02-05 Protein lipidation in health and disease Rebeca M. Mejias Estevez,William Fuller,2023-11-15

Electron Microscopy Methods and Protocols M. A. Nasser Hajibagheri,2008-02-02 Electron Microscopy Methods and Protocols is designed for the established researcher as a manual for extending knowledge of the field It is also for the newcomer who wishes to move into the field A wide range of applications for the examination of cells tissues biological macromolecules molecular structures and their interactions are discussed We have tried to gather together methods that we consider to be those most generally applicable to current research in both cell and molecular biology Each chapter contains a set of related practical protocols with examples provided by experts who have first hand knowledge of the techniques they describe The individual chapters are grouped according to similarities in their specimen preparation and methodology Methods are presented in detail in a step by step fashion using reproducible protocols the authors have personally checked During the last decade the scientific literature describing the use of colloidal gold as an immunocytochemical marker has increased at an exponential rate and this trend is expected to continue We have included a large number of variations on the immunogold labeling technique In both the negative staining and cryo chapters authors emphasize the immunological applications in order to correlate as fully as possible with the emphasis on immunogold labeling in the other chapters Electron Microscopy Methods and Protocols commences with the routine preparation of biological material for classical transmission electron microscopy involving tissue fixation embedding and sectioning Chap 1 RNA-Protein Interaction Protocols Susan R. Haynes,2008-02-03 The molecular characterization of RNA and its interactions with proteins is an important and exciting area of current research Organisms utilize a variety of RNA protein interactions to regulate the expression of their genes This is particularly true for eukaryotes since newly synthesized messenger RNA must be extensively modified and transported to the cytoplasm before it can be used for protein synthesis The realization that posttranscriptional processes are critical components of gene regulation has sparked an explosion of interest in both stable ribonucleoprotein RNP complexes and transient RNA protein interactions RNA is conformationally flexible and can adopt complex structures that provide diverse surfaces for interactions with proteins The fact that short RNA molecules aptamers see Chapter 16 can be selected to bind many different types of molecules is evidence of the structural variability of RNA RNA molecules are rarely entirely single or double stranded but usually contain multiple short duplexes interrupted by single stranded loops and bulges in some RNAs such as tRNAs the short duplexes stack on each other Further variability is generated by the presence of non Watson Crick base pairs modified nucleotides and more complex structures such as pseudoknots and triple strand interactions Complement Methods and Protocols B. Paul Morgan,2008-02-05 The complement system first described more than a century ago was for many years the ugly duckling of the immunology world but no more Complement in recent years has blossomed into a fascinating and fast moving field of immediate relevance to clinical scientists in fields as diverse as transplantation biology virology and inflammation Despite its emergence from the shadows complement retains an

unwarranted reputation for being difficult This impression derives in large part from the superficially complicated nomenclature a relic of the long and tortuous process of unraveling the system of naming components in order of discovery rather than in a systematic manner Once the barrier of nomenclature has been surmounted then the true simplicity of the system becomes apparent Complement comprises an activation system and a cytolytic system The former has diverged to focus on complement to distinct targets bacteria immune complexes and others so that texts now describe three activation pathways closely related to one another but each with some unique features The cytolytic pathway is the same regardless of the activation process and kills cells by creating pores in the membrane Complement plays an important role in killing bacteria and is essential for the proper handling of immune complexes Problems occur when complement is activated in an inappropriate manner the potent inflammation inducing products of the cascade then cause unwanted tissue damage and destruction

Bioconjugation Protocols Christof M. Niemeyer, 2008-02-04 There are a number of outstanding volumes that provide a comprehensive overview of bioconjugation techniques However many of the conventional approaches to the synthesis of chemically modified protein conjugates lack efficient means to control the stoichiometry of conjugation as well as the specific site of attachment of the conjugated moiety Moreover the recent developments in microarray technologies as well as in nanobiotechnology a novel field of research rapidly evolving at the crossroads of physics chemistry technology and materials science call for a summary of modern bioconjugation strategies to overcome the limitations of the classical approaches Bioconjugation Protocols Methods and Strategies is intended to provide an update of many of the classic techniques and also to introduce and summarize newer approaches that go beyond the pure biomedical applications of bioconjugation The purpose of Bioconjugation Protocols Methods and Strategies is therefore to provide instruction and inspiration for all those scientists confronting the challenges of semisynthesizing functional biomolecular reagents for a wide variety of applications ranging from novel biomedical diagnostics to therapeutics to biomaterials Part I contains seven protocols for the preparation of protein conjugates

Methods in Molecular Biology: Protein lipidation protocols John M. Walker, 1984

Developmental Biology Protocols Rocky S. Tuan, Cecilia W. Lo, 2008-02-05 Developmental biology is one of the most exciting and fast growing fields today In part this is so because the subject matter deals with the innately fascinating biological events changes in form structure and function of the organism The other reason for much of the excitement in developmental biology is that the field has truly become the unifying melting pot of biology and provides a framework that integrates anatomy physiology genetics biochemistry and cellular and molecular biology as well as evolutionary biology No longer is the study of embryonic development merely embryology In fact developmental biology has produced important paradigms for both basic and clinical biomedical sciences alike Although modern developmental biology has its roots in experimental embryology and the even more classical chemical embryology the recent explosive and remarkable advances in developmental biology are critically linked to the advent of the cellular and molecular biology

revolution The impressive arsenal of experimental and analytical tools derived from cell and molecular biology which promise to continue to expand together with the exponentially developing sophistication in functional imaging and information technologies guarantee that the study of the developing embryo will contribute one of the most captivating areas of biological research in the next millennium

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

Chemokine Protocols Amanda E.I. Proudfoot, Timothy N.C. Wells, Christine Power, 2008-02-05 The chemokines family of small proteins are involved in numerous biological processes ranging from hematopoiesis angiogenesis and basal leukocyte trafficking to the extravasation and tissue infiltration of leukocytes in response to inflammatory agents tissue damage and bacterial or viral infection Chemokines exert their effects through a family of seven G protein coupled transmembrane receptors Worldwide interest in the chemokine field surged dramatically early in 1996 with the finding that certain chemokine receptors were the elusive coreceptors required along with CD4 for HIV infection Today though over 40 human chemokines have been described the number of chemokine receptors lags behind only 17 human chemokine receptors have been identified so far What has emerged over the years is that most chemokine receptors bind several distinct ligands and indeed the majority of chemokines are able to bind to multiple chemokine receptors explaining to some extent the apparent disparity in the numbers of chemokines and receptors Yet in spite of the apparent redundancy in chemokine receptor interactions it is clear that in vivo spatial temporal and indeed cell and tissue specific expression of both chemokines and their receptors are important factors in determining the precise nature of cellular infiltrates in physiological and pathological processes

Flavoprotein Protocols Steven K. Chapman, Graeme A. Reid, 2008-02-03 As a scientist with an interest in proteins you will at some time in your career isolate an enzyme that turns out to be yellow or perhaps you already have Alternatively you may identify a polypeptide sequence that is related to known flavin containing proteins This may or may not be your first encounter with flavoproteins However even if you are an old hand in the field you may not have exploited the full range of experimental approaches applicable to the study of flavoproteins We hope that Flavoprotein Protocols will encourage you to do so In this volume we have sought to bring together a range of experimental methods of value to researchers with an interest in flavoproteins whether or not these

researchers have experience in this area A broad range of techniques from the everyday to the more specialized is described by scientists who are experts in their fields and who have extensive practical experience with flavoproteins The wide range of approaches from wet chemistry to dry computation has as a consequence demanded a range of formats Where appropriate particularly for analytical methods the protocol described is laid out in easy to follow steps In other cases e g the more advanced spectroscopies and computational methods it is far more apt to describe the general approach and relevance of the methods We hope this wide ranging approach will sow the seeds of many future collaborations between laboratories and further our knowledge and understanding of how flavoproteins work

T Cell Protocols Kelly P. Kearse, 2008-02-02 The purpose of T Cell Protocols Development and Activation is to collect a series of protocols particularly those that have been developed within the past few years to help investigators master new techniques or improve existing ones for the study of T cell Biology Invariably in putting together a book like this it is difficult to decide which methods to include and which to leave out To this end methods were selected from a variety of disciplines including cellular immunology biochemistry and molecular biology to try to provide something of interest for everyone who works on T cell development and activation I would like to mention that my primary reason for agreeing to put this book together is that when I was a graduate student I purchased a copy of Selected Methods in Cellular Immunology by Mishell and Shigii which proved a tremendous help in learning the basics of one and two dimensional gel techniques and other methods The cover has long since fallen off but it still remains one of my most valued reference books for the laboratory It is my hope that T Cell Protocols Development and Activation will prove similarly useful to current and future scientists wishing to learn new methods for exploring the development and activation of T cells

Mycotoxin Protocols Mary W. Trucksess, Albert E. Pohland, 2008-02-05 Mycotoxins produced by molds are common contaminants of many important crops including wheat corn rice and peanuts Some mycotoxins are found in fruits and vegetables These contaminants have a broad range of toxic effects including carcinogenicity neurotoxicity and reproductive and developmental toxicity The occurrence of mycotoxins in foods is an unavoidable worldwide problem About 80 countries have imposed regulatory limits to minimize human and animal exposure to mycotoxins Regulatory limits including international standards have tremendous economic impact and must be developed using science based risk assessments The purpose of Mycotoxin Protocols is to provide the scientific and technological basis for analytical methods for use in obtaining the exposure data needed for risk assessments Mycotoxin Protocols is divided into four sections which are interconnected The first section Chapters 1-5 describe the general techniques for mycotoxin analysis with emphasis on the importance of method validation based on statistical parameters sampling procedures for collecting a sample as representative as possible of a bulk lot the isolation of mycotoxins for use as analytical standards or for toxicological studies the evaluation of purity and preparation of standards and the detection and identification of impurities in isolated mycotoxins Sections 2-4 Chapters 6-19 describe the most current chromatographic and immunochemical methods for studies on the

major mycotoxins **Natural Killer Cell Protocols** Kerry S. Campbell, Marco Colonna, 2008-02-03 In *Natural Killer Cell Protocols Cellular and Molecular Methods* Kerry S Campbell and Marco Colonna have assembled a comprehensive collection of readily reproducible methods designed to study natural killer NK cells from the broadest variety of viewpoints These include not only classic techniques but also new approaches to standard methods newly evolved techniques that have become valuable for specific applications and unique models for manipulating and studying NK cells Among the advanced methods covered are those for in vitro transendothelial migration in vivo detection of cells migrating into tumors immunofluorescence staining of intracellular cytokines and in vitro NK cell development Valuable techniques for specific applications include vaccinia virus protein expression soluble KIR Fc fusions for HLA class I binding assays calcium mobilization in cell conjugates and identification of heterodimeric receptor complexes using cDNA library expression cloning No less important are accounts of such classic methods as hybrid resistance ADCC viral defense target cell cytotoxicity assays cloning and culturing tumor immunotherapy and generation of HLA class I transfected target cells *Natural Killer Cell Protocols Cellular and Molecular Methods* offers immunologists cancer researchers virologists and cell biologists today's most comprehensive collection of both established and cutting edge techniques methods that will contribute significantly to advancing our understanding of this fascinating and critically important class of cells

Chromatin Protocols Peter B. Becker, 2008-02-03 More than 40 years after the discovery of the nucleosome as the fundamental unit of chromatin the multifaceted problem of how variations in chromatin structure affect the activity of the eukaryotic genome has not been solved However during the past few years research on chromatin structure and function has gained considerable momentum and impressive progress has been made at the level of concept development as well as filling in crucial detail The structure of the nucleosome has been visualized at unprecedented resolution Powerful multisubunit enzymes have been identified that alter histone DNA interactions in ways that expose regulatory sequences to factors initiating and regulating such nuclear processes as transcription Though the importance of posttranslational modifications of histones notably their acetylation has long been known the finding that a number of bona fide regulators increase transcription by acetylating nucleosomes has lent new support to the old idea that the process of gene regulation is intimately related to the nature of the chromatin environment A wealth of nonhistone proteins contribute to a continuum of structures with distinct biochemical properties and varying degrees of DNA condensation Perhaps the most important conclusion from a large number of studies is a fresh appreciation of the dynamic nature of chromatin structure the built in flexibility providing the basis for regulation

Transforming Growth Factor-Beta Protocols Philip H. Howe, 2008-02-05 Philip H Howe and a group of well versed experimentalists present the first major volume a collection of indispensable classic and cutting edge TGF assays Described in great detail to ensure robust and successful results these readily reproducible techniques range from the growth inhibition assay for TGF to methods for monitoring its interactions with the mediating proteins Extensive notes discuss potential pitfalls and provide tips on how to

avoid failures and throughout emphasis is given to detailing those technical steps critical for experimental success that are often omitted in the primary literature Concise and highly practical Transforming Growth Factor Beta Protocols provides today s molecular and cell biologists both expert and novice with time tested methods for the identification and analysis of the signal transduction pathways by which TGF induces and modulates physiological behavior

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