

Methods in Molecular Biology™

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Glycoprotein Methods and Protocols

The Mucins

Edited by
Anthony P. Corfield



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Glycoprotein Methods And Protocols The Mucins Methods In Molecular Biology

John R. Crowther



Glycoprotein Methods And Protocols The Mucins Methods In Molecular Biology:

Glycoprotein Methods and Protocols Anthony P. Corfield, 2007-10-26 The mucins mucus glycoproteins have long been a complex corner of glycoprotein biology While dramatic advances in the separation structural analysis biosynthesis and degradation have marked the progress in general glycoprotein understanding the mucins have lagged behind The reasons for this lack of progress have always been clear and are only now being resolved The mucins are very large molecules they are difficult to separate from other molecules present in mucosal secretions or membranes they are often degraded owing to natural protective functions or to isolation methodology and their peptide and oligosaccharide structures are varied and complex Understanding these molecules has demanded progress in several major areas Isolation techniques that protect the intact mucins and allow dissociation from other adsorbed but discrete molecules needed to be developed and accepted by all researchers in the field Improved methods for the study of very large molecules with regard to their aggregation and polymerization were also needed Structural analysis of the peptide domains and the multitude of oligosaccharide chains was required for smaller sample sizes for multiple samples and in shorter time In view of these problems it is perhaps not surprising that the mucins have remained a dilemma of obvious biological importance and interest but very difficult to analyze

Methods in Molecular Biology: Glycoprotein methods and protocols : the mucins John M. Walker, 1984 *Mucins* Michael A. McGuckin, David J. Thornton, 2012-01-19 Epithelial mucins are large complex cell surface and secreted glycoproteins produced by mucosal epithelial cells In *Mucins Methods and Protocols* expert researchers in the field detail many of the methods which are now commonly used to study Mucins These include methods and techniques for the best approaches to analysing each specific area of mucin biochemistry physiology and biophysics before providing individual detailed experimental protocols together with troubleshooting and interpretation tips 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 key tips on troubleshooting and avoiding known pitfalls Authoritative and practical *Mucins Methods and Protocols* is designed to be a useful resource for those entering the mucin field and to facilitate those already studying mucins to broaden their experimental approaches to understanding mucosal biology

Glycoprotein Methods and Protocols Anthony P. Corfield, 2000-03-01 The mucins mucus glycoproteins have long been a complex corner of glycoprotein biology While dramatic advances in the separation structural analysis biosynthesis and degradation have marked the progress in general glycoprotein understanding the mucins have lagged behind The reasons for this lack of progress have always been clear and are only now being resolved The mucins are very large molecules they are difficult to separate from other molecules present in mucosal secretions or membranes they are often degraded owing to natural protective functions or to isolation methodology and their peptide and oligosaccharide structures are varied and complex Understanding these molecules has demanded progress in several major areas Isolation

techniques that protect the intact mucins and allow dissociation from other adsorbed but discrete molecules needed to be developed and accepted by all researchers in the field Improved methods for the study of very large molecules with regard to their aggregation and polymerization were also needed Structural analysis of the peptide domains and the multitude of oligosaccharide chains was required for smaller sample sizes for multiple samples and in shorter time In view of these problems it is perhaps not surprising that the mucins have remained a dilemma of obvious biological importance and interest but very difficult to analyze

Neuropeptide Y Protocols Ambikaipakan Balasubramaniam, 2008-02-05 The observation that neuropeptide Y NPY is the most abundant peptide present in the mammalian nervous system and the finding that it elicits the most powerful orexigenic signal have led to active investigations of the properties of the NPY family of hormones including peptide YY PYY and pancreatic polypeptide PP Nearly two decades of research have led to the identification of several NPY receptor subtypes and the development of useful receptor selective ligands Moreover these investigations have implicated NPY in the pathophysiology of a number of diseases including feeding disorders seizures memory loss anxiety depression and heart failure Vigorous efforts are therefore continuing not only to understand the biochemical aspects of NPY actions but also toward developing NPY based treatments for a variety of disorders To facilitate these efforts it was decided to produce the first handbook on NPY research techniques as part of the Methods in Molecular Biology Series In compiling *Neuropeptide Y Protocols* I have gathered contributions on techniques considered critical for the advancement of the NPY field from experts in various disciplines Each chapter starts with a brief introduction with Materials and Methods sections following The latter sections are presented in an easy to follow step by step format The last section of the chapter Notes highlights pitfalls and the maneuvers employed to overcome them This information not usually disseminated in standard research publications may prove extremely useful for investigators employing these techniques in NPY research

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

Extracellular Matrix Protocols Charles Streuli, Michael Grant, 2008-02-05 It is now widely accepted that much of the dynamic function of cells and tissues is regulated from outside the cell by the extracellular matrix In addition to its conventional role in providing a scaffold for building tissues the extracellular matrix acts as a directional highway for cellular movement and provides instructional information for promoting survival proliferation and differentiation Indeed the extracellular matrix is beginning to take a starring role in the choreography of cell and tissue function The diverse roles of the extracellular matrix are reflected in its highly complicated structure consisting of an ever increasing number of components Yet the mechanisms of extracellular matrix assembly and how they influence cell behavior are only just beginning to be understood In order to solve these problems new methodologies are of necessity being developed Many of these technologies are highly sophisticated and are currently available only in a handful of laboratories However we believe that they can readily be transported and established by other researchers Thus the purpose of Extracellular Matrix Protocols is to present some of these complicated techniques in a style that is relatively easy to reproduce

Connexin Methods and Protocols Roberto Bruzzone, Christian Giaume, 2008-02-05 Direct cell-cell communication is a common property of multicellular organisms that is achieved through membrane channels which are organized in gap junctions The protein subunits of these intercellular channels the connexins form a multigene family that has been investigated in great detail in recent years It has now become clear that in different tissues connexins speak several languages that control specific cellular functions This progress has been made possible by the availability of new molecular tools and the improvement of basic techniques for the study of membrane channels as well as by the use of genetic approaches to study protein function in vivo More important connexins have gained visibility because mutations in some connexin genes have been found to be linked to human genetic disorders Connexin Methods and Protocols presents in detail a collection of techniques currently used to study the cellular and molecular biology of connexins and their physiological properties The field of gap junctions and connexin research has always been characterized by a multidisciplinary approach combining morphology biochemistry biophysics and cellular and molecular biology This book provides a series of cutting edge protocols and includes a large spectrum of practical methods that are available to investigate the function of connexin channels Connexin Methods and Protocols is divided into three main parts

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

Developmental Biology Protocols Rocky S. Tuan, Cecilia W. Lo, 2000-01-21 This three volume set consisting of 142 chapters is intentionally broad in scope because of the nature of modern developmental biology

Cytoskeleton Methods and Protocols Ray H. Gavin, 2008-02-03 Over the past two decades experimental studies have solidified the interpretation of the cytoskeleton as a highly dynamic network of microtubules actin microfilaments intermediate filaments and myosin filaments Rather than a network of disparate fibers these polymers are often interconnected and display synergy which is the combined action of two or more cytoskeletal polymers to achieve a specific cellular structure or function Cross communication among cytoskeletal polymers is thought to be achieved through cytoskeletal polymer accessory proteins and molecular motors that bind two or more cytoskeletal polymers Development of the modern concept of the cytoskeleton is a direct outgrowth of advances in experimental tools and reagents that are available to cell and molecular biologists Technological advances and refinements in cell imaging have made it possible to selectively image a single cytoskeletal polymer and monitor its dynamics through the use of fluorescence probes in vitro and in vivo Two decades ago cytoskeletal research was limited to a few perturbation reagents that included colchicine and cytochalasin Today the perturbation arsenal has expanded to a highly selective group of reagents that includes Taxol nocodazole benomyl latrunculin jasplakinolide and such endogenous proteins as gelsolin These reagents enable the investigator to selectively perturb or destroy a cytoskeletal polymer while leaving other cytoskeletal polymers intact Site specific monoclonal antibodies that target a specific cytoskeletal polymer have proven to be highly selective affinity tools for cytoskeletal research

Immunotoxin Methods and Protocols Walter A. Hall, 2008-02-02 Immunotoxins represent a new class of human therapeutics that have widespread applications and a potential that has not yet been fully recognized since they were first conceived of by Paul Ehrlich in 1906 The majority of advances in the development and implementation of immunotoxins has occurred over the last 20 years The reasons for this use of immunotoxins in basic science and clinical research are the powerful concurrent advances in genetic engineering and receptor physiology Recombinant technology has allowed investigators to produce sufficient quantities of a homogeneous compound that allows clinical trials to be performed The identification of specific receptors on malignant cell types has enabled scientists to generate immunotoxins that have had positive results in clinical trials As more cellular targets are identified in coming years additional trials will be conducted in different disease states affecting still larger patient populations Modulation of the immune system to decrease the humoral response to immunotoxins may improve their overall efficacy As increasingly more effective compounds are generated it will be necessary to decrease the local and systemic toxicity associated with these agents and methods for doing so are presently being developed The work presented in Immunotoxin Methods and

Protocols focuses on three specific areas of immunotoxin investigation that are being conducted by experts throughout the world The first section describes the construction and development of a variety of immunotoxins

DNA Topoisomerase Protocols Neil Osheroff, Mary-Ann Bjornsti, 2008-02-05 Beginning with the Escherichia coli protein or bacterial DNA topoisomerase I an ever increasing number of enzymes have been identified that catalyze changes in the linkage of DNA strands DNA topoisomerases are ubiquitous in nature and have been shown to play critical roles in most processes involving DNA including DNA replication transcription and recombination These enzymes further constitute the cellular targets of a number of clinically important antibacterial and anticancer agents Thus further studies of DNA topology and DNA topoisomerases are critical to advance our understanding of the basic biological processes required for cell cycle progression cell division genomic stability and development In addition these studies will continue to provide critical insights into the cytotoxic action of drugs that target DNA topoisomerases Such mechanistic studies have already played an important role in the development and clinical application of antimicrobial and chemotherapeutic agents The two volumes of DNA Topoisomerase Protocols are designed to help new and established researchers investigate all aspects of DNA topology and the function of these enzymes The chapters are written by prominent investigators in the field and provide detailed background information and step by step experimental protocols The topics covered in Part I DNA Topology and Enzymes range from detailed methods to analyze various aspects of DNA structure from linking number knotting unknotting site specific recombination and decatenation to the overexpression and purification of bacterial and eukaryotic DNA topoisomerases from a variety of cell systems and tissues

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

Plant Hormone Protocols Gregory A. Tucker, Jeremy A. Roberts, 2008-02-04 Established investigators from

around the world describe in step by step detail their best techniques for the study of plant hormones and their regulatory activities These state of the art methods include contemporary approaches to identifying the biosynthetic pathways of plant hormones monitoring their levels characterizing the receptors with which they interact and analyzing the signaling systems by which they exert their effects Comprehensive and fully detailed for reproducible laboratory success *Plant Hormone Protocols* offers plant biologists an indispensable compendium of today s most powerful methods and strategies to studying plant hormones their regulation and their activities

The ELISA Guidebook John R. Crowther, 2008-02-04 John R Crowther provides today s premier practical guide to the understanding and application of ELISA Updating and greatly expanding his widely appreciated earlier publication *ELISA Theory and Practice* 1995 this important work introduces chapters on such major new topics as checkerboard titrations quality control of testing kit production and control novel monoclonal antibodies validation of assays statistical requirements for data examination and epidemiological considerations With its numerous worked examples detailed instructions and extensive illustrations *The ELISA Guidebook* offers a powerful synthesis of all the basic concepts and practical experimental details investigators need to understand develop and apply the new ELISA methodology successfully in day to day basic and clinical research

Fortschritte der Chemie organischer Naturstoffe / Progress in the Chemistry of Organic Natural Products 85 , 2012-12-06 The volumes of this classic series now referred to simply as Zechmeister after its founder L Zechmeister have appeared under the Springer Imprint ever since the series inauguration in 1938 The volumes contain contributions on various topics related to the origin distribution chemistry synthesis biochemistry function or use of various classes of naturally occurring substances ranging from small molecules to biopolymers Each contribution is written by a recognized authority in his field and provides a comprehensive and up to date review of the topic in question Addressed to biologists technologists and chemists alike the series can be used by the expert as a source of information and literature citations and by the non expert as a means of orientation in a rapidly developing discipline

Bioinformatics Methods and Protocols Stephen Misener, Stephen A. Krawetz, 2008-02-02 Computers have become an essential component of modern biology They help to manage the vast and increasing amount of biological data and continue to play an integral role in the discovery of new biological relationships This in silico approach to biology has helped to reshape the modern biological sciences With the biological revolution now among us it is imperative that each scientist develop and hone today s bioinformatics skills if only at a rudimentary level *Bioinformatics Methods and Protocols* was conceived as part of the *Methods in Molecular Biology* series to meet this challenge and to provide the experienced user with useful tips and an up to date overview of current developments It builds upon the foundation that was provided in the two volume set published in 1994 entitled *Computer Analysis of Sequence Data* We divided *Bioinformatics Methods and Protocols* into five parts including a thorough survey of the basic sequence analysis software packages that are available at most institutions as well as the design and implementation of an essential introductory Bioinformatics course In addition we

included sections describing specialized noncommercial software databases and other resources available as part of the World Wide Web and a stimulating discussion of some of the computational challenges biologists now face and likely future solutions

Mass Spectrometry of Proteins and Peptides John R. Chapman, 2008-02-05 Little more than three years down the line and I am already writing the Preface to a second volume to follow *Protein and Peptide Analysis by Mass*. What has happened in between these times to make this second venture worthwhile? New types of mass spectrometric instrumentation have appeared so that new techniques have become possible and existing techniques have become much more feasible. More particularly, however, the newer ionization techniques introduced for the analysis of high molecular weight materials have now been thoroughly used and studied. As a result, there has been an enormous improvement in the associated sample handling technology so that these methods are now routinely applied to much smaller sample amounts as well as to more intractable samples. Again, this particular community of mass spectrometry users has both increased in number and diversified. And riding this wave of acceptance, leaders in the field have set their sights on more complex problems: molecular interaction, ion structures, quantitation, and kinetics are just a few of the newer areas reported in *Mass Spectrometry of Proteins and Peptides*. As with the first volume, one purpose of this collection, *Mass Spectrometry of Proteins and Peptides*, is to show the reader what can be done by the application of mass spectrometry and perhaps even to encourage the reader to venture down new paths.

DNA-Protein Interactions Tom Moss, 2008-02-02 Dr Tom Moss assembles the new standard collection of cutting edge techniques to identify key protein-DNA interactions and define their components, their manner of interaction, and their manner of function both in the cell and in the test tube. The techniques span a wide range from factor identification to atomic detail and include multiple DNA footprinting analyses including *in vivo* strategies, gel shift, EMSA optimization, SELEX, surface plasmon resonance, site-specific DNA-protein crosslinking, and UV laser crosslinking. *Comprehensive and Broad-Ranging DNA-Protein Interactions: Principles and Protocols*, 2nd Edition, offers a stellar array of over 100 up-to-date and readily reproducible techniques that biochemists and molecular, cellular, and developmental biologists can use successfully today to understand DNA-protein interactions.

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