**VOLUME 141** 

# Plant Hormone Protocols

Edited by

Gregory A. Tucker Jeremy A. Roberts



# Plant Hormone Protocols Methods In Molecular Biology Vol 141

**Minjie Lin** 

#### Plant Hormone Protocols Methods In Molecular Biology Vol 141:

**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 The ELISA Guidebook John R. Crowther, 2008-02-04 John R Crowther provides their regulation and their activities 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 **Molecular Methods in Developmental Biology** Matt Guille, 2008-02-03 The process whereby a single cell the fertilized egg develops into an adult has fascinated for centuries Great progress in understanding that process hever has been made in the last two decades when the techniques of molecular biology have become available to developmental biologists By applying these techniques the exact nature of many of the interactions responsible for forming the body pattern are now being revealed in detail Such studies are a large and it seems ever expanding part of most life science groups It is at newcomers to this field that this book is primarily aimed A number of different plants and animals serve as common model org isms for developmental studies In Molecular Methods in Developmental Bi ogy Xenopus and Zebrafish a range of the molecular methods applicable to two of these organisms are described these are the South African clawed frog Xenopus laevis and the zebrafish Brachydanio rerio The embryos of both of these species develop rapidly and externally making them particularly suited to investigations of early vertebrate development However both Xenopus and zebrafish have their own advantages and disadvantages Xenopus have large robust embryos that can be manipulated surgically with ease but their pseudotetraploidy and long generation time make them unsuitable candidates for genetics This disadvantage may soon be overcome by using the diploid Xenopus tropicalis and early experiments are already underway The transp ent embryos of zebrafish render them well suited for in situ hybridization and immunohistochemistry and good for observing mutations in genetic screens **Proteoglycan Protocols** Renato V. Iozzo,2008-02-02 Proteoglycans are some of the most elaborate macromolecules of mammalian and lower organisms The

covalent attachment of at least five types of glycosami glycan side chains to more than forty individual protein cores makes these molecules quite complex and endows them with a multitude of biological functions Proteoglycan Protocols offers a comprehensive and up to date collection of prepa tive and analytical methods for the in depth analysis of proteoglycans Featuring st by step detailed protocols this book will enable both novice and experienced researchers to isolate intact proteoglycans from tissues and cultured cells to establish the composition of their carbohydrate moieties to generate strategies for prokaryotic and eukaryotic expression to utilize methods for the suppression of specific proteoglycan gene expression and for the detection of mutant cells and degradation products and to study specific interactions between proteoglycans and extracellular matrix proteins as well as growth factors and their receptors The readers will find concise yet comprehensive techniques carefully drafted by leading experts in the field Each chapter commences with a general Introduction followed by a detailed Materials section and an easy to follow Methods section An asset of each chapter is the extensive notation that includes troubleshooting tips and practical considerations that are often lacking in formal methodology papers The reader will find this section most valuable because it is clearly provided by experienced scientists who have first hand knowledge of the techniques they outline In addition most of the chapters are well illustrated with examples of typical data generated with each method Biology and Biotechnology of the Plant Hormone Ethylene III Miguel Vendrell, 2003 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 te niques 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 c bining 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 fu tion of connexin channels Connexin Methods and Protocols is divided into three main parts Neurotrophin Protocols Robert A. Rush, 2008-02-03 The past decade has seen an extraordinary growth in research interest in neurotrophic factors and the study of the neurotrophin family has led this activity Nevertheless this area of research has often struggled as a result of techniques that were either inadequate or just emerging from other research fields and disciplines Neurotrophin Protocols

has brought together many leaders in the neurotrophin field who detail their special expertise in a wide variety of techniques Though most procedures are valid across many diff ent fields of research some of those described here have been developed to address particular issues within the neurotrophic factor field. The protocols cover a broad range of biochemical histological and biological techniques that are often required by the modern laboratory However all have been written with sufficient detail to allow any laboratory to achieve proficiency without need of reference to other texts Neurotrophin Protocols is divided into four sections dealing with p tein RNA recombinant and in vivo techniques Protein techniques have in general been less successfully employed than those dealing with RNA or DNA However procedures that achieve localization and quantification of the neurotrophins are now being used more extensively Their inclusion here should assist further studies at the protein level Transgenic cell lines and animals are commonplace in the scientific research literature but their inc sion in several chapters in this book provide some novel uses that are not readily available elsewhere **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 c pound 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 sociated with these agents and methods for doing so are presently being veloped. 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

**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 Seguence 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 implemention 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 stimul ing discussion of some of the computational challenges biologists now face and likely future 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 org ism 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 mole lar biology as well as evolutionary biology No longer is the study of embryonic development merely embryology In fact development biology has produced portant paradigms for both basic and clinical biomedical sciences alike Although modern developmental biology has its roots in experimental emb ology 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 expe mental and analytical tools derived from cell and molecular biology which promise to continue to expand together with the exponentially developing sophistication in fu tional imaging and information technologies guarantee that the study of the devel ing embryo will contribute one of the most captivating areas of biological research in the next millennium Nuclease Methods and Protocols Catherine H. Schein, 2008-02-03 Nucleases enzymes that restructure or degrade nucleic acid polymers are vital to the control of every area of metabolism They range from housekeeping enzymes with broad substrate ranges to extremely specific tools 1 Many types of nucleases are used in lab protocols and their commercial and clinical uses are expanding The purpose of Nuclease Methods and Protocols is to introduce the reader to some we characterized protein nucleases and the methods used to determine their activity structure interaction with other molecules and physiological role Each chapter begins with a mini review on a specific nuclease or a nuclease related theme Although many chapters cover several topics they were arbitrarily divided into five parts Part I Characterizing Nuclease Activity includes protocols and assays to determine general processive distributive or specific mechanisms Methods to assay nuclease products identify cloned nucleases and determine their physiological role are also included here Part II Inhibitors and Activators of Nucleases summarizes assays for measuring the effects of other proteins and small molecules Many of these inhibitors have clinical relevance Part III Relating Nuclease Structure and Function provides an overview of methods to determine or model the 3 D structure of nucleases and their complexes with substrates and inhibitors A 3 D structure can greatly aid the rational design of nucleases and inhibitors for specific purposes Part IV Nucleases in the Clinic summarizes

assays and protocols suitable for use with t sues and for nuclease based therapeutics Adipose Tissue Protocols Gérard Ailhaud, 2008-02-03 Adipose tissue is recognized to be exquisitely sensitive to hormone action and is also now recognized as a secretory and endocrine organ required for reproduction and good health Adipocytes are smart cells able within the tissue to communicate with surrounding cells but also with various organs particularly via leptin acting on the central nervous system Brown adipose tissue BAT and white adipose tissue WAT are known to be distinct tissues whereas the heterogeneity of WAT depots is well est lished Unfortunately excess WAT leads to obesity which is the most common health problem in industrialized countries Therefore from both a scientific and a technical point of view the time has come to create a survey of adipose tissues and their neglected adipocytes In Adipose Tissue Protocols I have attempted to gather together chapters from all areas of adipose tissue research from in vivo to in vitro studies and to provide methods covering a wide variety of techniques including the choice of adipose tissue depot and of morphological techniques for the study of BAT and WAT the isolation subcellular fractionation and transfection of adipocytes where the low density of these cells must be taken into account assays of nutrient and ion fluxes and the metabolic aspects of nutrient uptake assays of lipid related enzymes biopsies and quantification of lipid related mRNAs cultures of adipose precursor cells from WAT and BAT of various species including human tissue measurements of adipose secretory products and assessment of WAT metabolism in vivo

Cytoskeleton Methods and Protocols Ray H. Gavin, 2008-02-03 Over the past two decades experimental studies have solidified the int pretation 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 commu cation 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 o growth 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 po mer 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

**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 interc nected 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 impu ties in isolated mycotoxins Sections 2 4 Chapters 6 19 describe the most current chromatographic and immunochemical methods for studies on the major mycotoxins Amino Acid Analysis Protocols Catherine Cooper, 2008-02-05 A collection of classic and cutting edge techniques of high utility in answering specific biological questions about amino acids Common methods include those based on HPLC or gas chromatography separation and analysis after precolumn derivatization New techniques based on capillary electrophoresis separation high performance anion exchange chromatography and mass spectrometry are also presented Each method is described in step by step detail to ensure successful experimental results and emphasizes sample preparation particularly the collection and storage of bodily fluids Up to date and highly practical Amino Acid Analysis Protocols offers analytical and clinical chemists as well as a broad range of biological and biomedical investigators a rich compendium of laboratory tools for the productive analysis of both common and uncommon amino acids **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 Calpain **Methods and Protocols** John S. Elce, 2008-02-05 The purpose of Calpain Methods and Protocols is guite straightf ward it is to present the actual experimental methods used in many different laboratories for the study of calpain It will provide the vital experimental detail and the discussion of possible pitfalls for which the standard journals no longer provide space This will make it as easy as possible for investi tors interested in calpain to adopt established methods without repeating old

mistakes and to adapt and apply these methods in novel approaches to the many outstanding calpain questions These questions range from purely biochemical problems of protein structure and enzyme regulation at the molecular level through large areas of cell biology to applied and clinical aspects of calpain function in human dease Within this panoply of topics a wide range of investigators will find many fascinating and as yet unanswered questions about calpain Calpain Methods and Protocols will provide instant access to many essential te niques while saving them the time and effort involved in developing a new method In addition to questions relating to the normal physiological roles of the calpains there is considerable evidence that inappropriate calpain activity may have pathological effects in many tissues for example following ischemia This provides a major stimulus for the development of specific calpain inhi tors for therapeutic purposes and for the development of methods to evaluate such inhibitors 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 syst atic 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 mune complexes and others so that texts now describe three activation pa ways 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 Gene Knockout Protocols Martin J. Tymms, Ismail Kola, 2008-02-03 As the major task of sequencing the human genome is near completion and full complement of human genes are catalogued attention will be focused on the ultimate goal to understand the normal biological functions of these genes and how alterations lead to disease states In this task there is a severe limitation in working with human material but the mouse has been adopted as the favored animal model because of the available genetic resources and the highly conserved gene conservation linkage organization In just of ten years since the first gene targeting experiments were p formed in embryonic stem ES cells and mutations transmitted through the mouse germline more than a thousand mouse strains have been created These achievements have been made possible by pioneering work that showed that ES cells derived from preimplantation mouse embryos could be cultured for prolonged periods without differentiation in culture and that homologous rec bination between targeting constructs and endogenous DNA occurred at a f quency sufficient for recombinants to be isolated In the next few years the mouse genome will be systematically altered and the techniques for achi ing manipulations are constantly being streamlined and improved 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 ext sive 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 q 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 tween laboratories and further our knowledge and understanding of how f voproteins work

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