Cytokines and Colony Stimulating Factors
Cytokines and Colony Stimulating Factors

Methods and Protocols

Edited by

Dieter Körholz

and

Wieland Kiess

Department of Pediatrics, University of Leipzig,
Leipzig, Germany

Humana Press  Totowa, New Jersey
Preface

The immune system is a complex network in which different cell types and soluble factors interact to efficiently eliminate various kinds of microorganisms as well as aberrant cell clones. The roots of immunologic investigations reach far into the past. In 430 BC, Thucydides reported that survivors of the plague did not present a second time with similar symptoms. The first report of a successful immunotherapy was made by Edward Jenner in 1798 who found a protective effect of cowpox vaccination against human pox. Since then, much knowledge has been accumulated; today, investigations of the molecular mechanisms of immune regulation are of central research interest. The novel insights into gene polymorphisms and gene regulation gathered from this work has improved our knowledge of individual immune reactions and risk factors in overcoming infections. Strategies to use the immune system for cancer treatment have been propelled by the discovery of divergent immunoregulatory cytokines and the introduction of new gene therapy strategies to modify immune responses. Recently, the discovery of various dendritic cells has focused attention on these cell types as central elements of the immune response and to the possibility of dendritic cell expansion, maturation, and consecutive stimulation with immunoreactive tumor-specific peptides. Similarly, methods for ex vivo expansion of various stem cell-derived cell types have led to an improved therapeutic management of various benign and malignant diseases. However, hope for greater therapeutic success in the clinical setting, and therefore patient benefits, has been quite limited with these methods. In the field of immunoregulation, enormous effort is still required to increase fundamental understanding and the therapeutic benefit. Only basic science and further research will provide for means to achieve this goal.

_Cytokines and Colony Stimulating Factors: Methods and Protocols_ is intended to promote all the present knowledge and understanding generated by the many novel technologies described here in the hope of spreading awareness of its useful applications. Our book aims to address the needs of novice investigators hoping to work in this field who need profound and practical information to get started, as well as seasoned investigators seeking to extend their technical range. The book not only provides detailed descriptions of methods, but also includes a section on troubleshooting in each chapter indicated in the Notes section. With the spreading use of these methods and the growing activity in the field, many qualified scientists can work on the therapeutic benefit of these studies for future patients suffering from these diseases.

_Dieter Körholz_
_Wieland Kiess_
Contents

Preface .......................................................................................................................... v
Contributors ................................................................................................................ xi

PART I. INTRODUCTION
1 Primary Immunodeficiencies Caused by Defects of Cytokines and Cytokine Receptors
   Volker Wahn .................................................................................................................. 3

PART II. DETECTION ASSAYS FOR CYTOKINES AND GROWTH FACTORS
2 Intracellular Detection of T-Cell Cytokines: Differentiation of TH1 and TH2 Cells
   Ursula Banning and Dieter Körholz ........................................................................... 15
3 Analysis of Cytokine Profiles in Human Skin
   Jean Krutmann, Renz Mang, and Susanne Grether-Beck ............................................ 23
4 Intracytoplasmic Detection of Proinflammatory Cytokines and Chemokines in Monocytes by Flow Cytometry
   Christian Schultz ........................................................................................................ 29
5 Flow-Cytometric Immune Function Methodology for Human Peripheral Blood Dendritic Cells
   Kerstin Willmann ........................................................................................................... 41
6 Evaluation of the Frequency of Virus-Specific CD8+ T Cells by Cytokine Flow Cytometry
   Bodo Hoffmeister, Felix Kiecker, Ingolf Surel, Elham Khatamzas, Volker Schuster, Hans-Dieter Volk, and Florian Kern ................. 59
7 Detection of Common Cytokine and Colony Stimulating Factor Gene Polymorphisms
   Thomas Lehrnbecher and Stephen J. Chanock ............................................................... 71
8 Apoptosis Induction by TRAIL
   Angelika Eggert, Hauke Sieverts, and Henning Walczak ........................................... 95
9 Angiogenic Cytokines: Quantitative and Functional Analysis
   Angelika Eggert and John H. Maris ............................................................................. 117
PART III. PROTOCOLS ON GENE THERAPY

10 Experimental Strategies for Combined Suicide and Immune Cancer Gene Therapy: An Overview
   Christof M. Kramm, Tim Niehues, and Nikolai G. Rainov .......... 137

11 Gene Therapy with Plasmids Encoding Cytokine- or Cytokine Receptor–IgG Chimeric Proteins
   Ciriac A. Piccirillo and Gérard J. Prud’homme ......................... 153

12 Interferon-β Gene Therapy for the Treatment of Arthritis
   Yuti Chernajovsky, Hanna Dreja, Kostas Triantaphyllopoulos, and David Gould ........................................... 171

13 Gene Gun-Based In Vivo Gene Transfer: Application to DNA Vaccination
   Tohru Sakai and Kunisuke Himeno ........................................... 181

14 Purification of the Eukaryotic Heat-Shock Proteins Hsp70 and gp96
   Arne von Bonin, Solveig H. Moré, and Minka Breloer ................ 193

15 Genetic Engineering of a Recombinant Fusion Protein Possessing an Antitumor Antibody Fragment and a TNF-α Moiety
   Jim Xiang and John R. Gordon ............................................... 201

16 Genetic Engineering of Dendritic Cells by Adenovirus-Mediated TNF-α Gene Transfer
   Jim Xiang and Josh Wu ..................................................... 213

17 Retroviral Transfer of T-Cell Receptor Genes into Human Peripheral Blood Lymphocytes
   Timothy M. Clay and Michael I. Nishimura ............................... 227

18 Gene Transfer to Articular Chondrocytes with Recombinant Adenovirus
   Glyn D. Palmer, Elvire Gouze, Jean-Noel Gouze, Oliver B. Betz, Christopher H. Evans, and Steven C. Ghivizzani .......... 235

19 Gene Therapy of X-Linked Severe Combined Immunodeficiency
   Salima Hacein-Bey-Abina, Genevieve de Saint Basile, and Marina Cavazzana-Calvo ......... 247

20 Gene Transfer for Generation of Tumor and Leukemia Vaccines
   Dagmar Dilloo and Andree Zibert ........................................... 261

21 Cytokine Gene Delivery into the Central Nervous System Using Intrathecaley Injected Nonreplicative Viral Vectors
   Roberto Furlan, Stefano Pluchino, Peggy C. Marconi, and Gianvito Martino .................. 279

PART IV. EX VIVO CELL EXPANSION

22 In Vitro Regulation of Colony Stimulating Factor-Mediated Hematopoiesis in Healthy Individuals and Patients with Different Types of Myeloproliferative Disease
   Thomas Vraetz, Peter D. Emanuel, and Charlotte M. Niemeyer ....... 293
Contents

23 In Vitro Generation of Dendritic Cells from Cord Blood CD34+ Hematopoietic Progenitors Cells
Bruno Canque, Michele Rosenzwajg, and Jean Claude Gluckman ....................................................... 311

24 Generation of Transgenic T Cells from Human CD34+ Cord Blood Cells
Bruno Verhasselt, Evelien Naessens, and Veronique Stove ...... 327

25 Ex Vivo Expansion of Umbilical Cord Blood Cells on Feeder Layers
Martin Bornhäuser ............................................................................................................................. 341

26 Ex Vivo Production of Cord Blood CD34+ Derived Myeloid Precursors After Serum-Free Static Culture
Sergio Querol ................................................................................................................................. 351

27 Ex Vivo Expansion of Hematopoietic Stem Cells
Beatriz Albella, José Carlos Segovia, Guillermo Guenechea, and Juan Antonio Bueren ............................................................ 363

28 Large-Scale Ex Vivo Expansion of Human Megakaryocytes for Clinical Use
Phil Lefebvre and Isaac Cohen .................................................................................. 375

29 Cytokine-Mediated Expansion of Human NOD–SCID-Repopulating Cells
Kohichiro Tsuji, Takahiro Ueda, and Yasuhiro Ebihara ........... 387

PART V. MATURATION AND DIFFERENTIATION OF DENDRITIC CELLS

30 Differentiation of Human Antigen-Presenting Dendritic Cells from CD34+ Hematopoietic Stem Cells In Vitro
Xin-Sheng Ju and Martin Zenke ............................................................... 399

31 Dendritic Cell Development from Mobilized Peripheral Blood CD34+ Cells
Deanne M. R. Lathers, Nicholas Achille, and M. Rita I. Young ...... 409

32 In Vitro Maturation of Dendritic Cells from Blood Progenitors
Lina Matera and Alessandra Galetto ................................................................. 417

33 Generation of Human Type 1- and Type 2-Polarized Dendritic Cells from Peripheral Blood
Pawel Kalinski, Pedro Vieira, Joost H. N. Schuitemaker, Quan Cai, and Martien Kapsenberg ................................. 427

34 Preparation of Human Dendritic Cells for Tumor Vaccination
Michael R. Shurin ............................................................................................... 437

35 Generation of Leukemic Dendritic Cells from Patients with Acute Myeloid Leukemia
Mohamad Mohty, Béatrice Gaugler, and Daniel Olive .................. 463

Index ............................................................................................................ 473
Contributors

NICHOLAS ACHILLE • Department of Pathology, Loyola University Stritch School of Medicine, Maywood, IL
BEATRIZ ALBELLA • Hematopoiesis Project, CIEMAT, Madrid, Spain
URSULA BANNING • Division of Pediatric Hematology and Oncology, Department of Pediatrics, University of Leipzig, Germany
OLIVER B. BETZ • Center for Molecular Orthopaedics, Harvard Medical School, Boston, MA
MARTIN BORNHÄUSER • Medical Clinic I, Carl Gustav Carus University, Dresden, Germany
MINKA BRELOER • Division of Medical Microbiology and Immunology, Bernhard-Nocht-Institute for Tropical Medicine, Hamburg, Germany
JUAN ANTONIO BUEREN • Hematopoiesis Project, CIEMAT, Madrid, Spain
QUAN CAI • Department of Surgery, University of Pittsburgh, PA
BRUNO CANQUE • Laboratoire d’Immunologie, Institut National de la Santé et de la Recherche Médicale, Paris, France
MARINA CAVAZZANA-CALVO • Gene and Cell Therapy Unit, Institut National de la Santé et de la Recherche Médicale, Hôpital Necker Enfants Malades, Paris, France
STEPHEN J. CHANOCK • Immunocompromised Host Section, Pediatric Oncology Branch, National Cancer Institute, National Institutes of Health, Gaithersburg, MD
YUTI CHERNAJOVSKY • Bone and Joint Research Unit, William Harvey Research Institute, Barts and Royal London Medical School, Queen Mary, University of London, UK
TIMOTHY M. CLAY • Program in Molecular Therapeutics, Department of Surgery, Duke University Medical Center, Durham, NC
ISAAC COHEN • Department of Cell and Molecular Biology, Northwestern University, Chicago, IL
GENEVIEVE DE SAINT BASILE • Institut National de la Santé et de la Recherche Médicale, Hôpital Necker Enfants Malades, Paris, France
Contributors

Dagmar Dilloo • Department of Pediatric Hematology and Oncology, University Hospital, Heinrich-Heine-University, Düsseldorf, Germany

Hanna Dreja • Bone and Joint Research Unit, William Harvey Research Institute, Barts and Royal London Medical School, Queen Mary, University of London, UK

Yasuhiro Ebihara • Division of Cellular Therapy, Advanced Clinical Research Center, Institute of Medical Science, University of Tokyo, Japan

Angelika Eggert • Department of Hematology/Oncology, University Children’s Hospital of Essen, Essen, Germany

Peter D. Emanuel • Department of Medicine, Comprehensive Cancer Center, University of Alabama, Birmingham, AL

Christopher H. Evans • Center for Molecular Orthopaedics, Harvard Medical School, Boston, MA

Roberto Furlan • Neuroimmunology Unit, Department of Neuroscience, San Raffaele Scientific Institute - DIBIT, Milan, Italy

Alessandra Galetto • Department of Oncological Surgery, University of Turin, Italy

Beatrice Gaugler • Laboratoire d’Immunologie des Tumeurs, Institut Paoli-Calmettes, Université de la Méditerranée, Institut National de la Santé et de la Recherche Médicale, Marseille, France

Steven C. Ghivizzani • Center for Molecular Orthopaedics, Harvard Medical School, Boston, MA

Jean Claude Gluckman • Laboratoire d’Immunologie, Institut National de la Santé et de la Recherche Médicale, Paris, France

John R. Gordon • Department of Veterinary Microbiology, Western College of Veterinary Medicine, University of Saskatchewan, Saskatoon, Canada

David Gould • Bone and Joint Research Unit, William Harvey Research Institute, Barts and Royal London Medical School, Queen Mary, University of London, UK

Elvire Gouze • Molecular Orthopedics, Harvard Medical School, Boston, MA

Jean-Noel Gouze • Center for Molecular Orthopaedics, Harvard Medical School, Boston, MA, and Electrical Engineering and Computer Sciences Laboratory, Massachusetts Institute of Technology, Cambridge, MA

Susanne Grether-Beck • Institut für Umweltmedizinische Forschung (IUF), Heinrich-Heine-University, Düsseldorf, Germany

Guillermo Guenechea • Hematopoiesis Project, CIEMAT, Madrid, Spain

Salima Hacein-Bey-Arbi • Gene and Cell Therapy Unit, Institut National de la Santé et de la Recherche Médicale, Hôpital Necker Enfants Malades, Paris, France
Contributors

KUNISuke HIMENO • Department of Parasitology and Immunology, Kyushu University Graduate School of Medicine, Fukuoka, Japan

BODO HOFFMEISTER • Institute of Medical Immunology, Charité, Humboldt-University, Berlin, Germany

XIN-SHENG JU • Max-Delbrück-Center for Molecular Medicine Berlin, Germany

Pawel KALINSKI • Department of Surgery, University of Pittsburgh, PA

Martien KAPSENBerg • Department of Cell Biology and Histology, Department of Dermatology, Academic Medical Center, University of Amsterdam, Netherlands

FLORIAN KERN • Institute of Medical Immunology, Charité, Humboldt-University, Berlin, Germany

Elham KHATAMZAS • Institute of Medical Immunology, Charité, Humboldt-University, Berlin, Germany

FELIX KIECKER • Institute of Medical Immunology, Charité, Humboldt-University, Berlin, Germany

WIELAND KIESS • Department of Pediatrics, University of Leipzig, Germany

Dieter Körholz • Division of Pediatric Hematology and Oncology, Department of Pediatrics, University of Leipzig, Germany

Christof M. Kramm • Department of Pediatric Hematology and Oncology, University Hospital, Heinrich-Heine-University, Düsseldorf, Germany

JEAN KRUTMANN • Institut für Umweltmedizinische Forschung (IUF), Heinrich-Heine-University, Düsseldorf, Germany

Deanne M. R. Lathers • Research Service, Hines Veterans Affairs Hospital, Hines, IL, and Departments of Pathology and Otolaryngology, Loyola University Strich School of Medicine, Maywood, IL

Phil LEFEBVRE • Department of Medicine, Northwestern University, Chicago, IL

Thomas LEHRNBECHER • Department for Pediatric Hematology and Oncology, Children’s Hospital III, Johann Wolfgang Goethe University, Frankfurt/Main, Germany

REnz MANG • Clinical and Experimental Photodermatology, Department of Dermatology, Heinrich-Heine-University, Düsseldorf, Germany

Peggy C. MARCONI • Section of Microbiology, Department of Clinical and Experimental Medicine, University of Ferrara, Italy

John H. MARIS • Division of Oncology, Children’s Hospital of Philadelphia, Philadelphia, PA

Gianvito MARTINO • Neuroimmunology Unit, Department of Neuroscience, San Raffaele Scientific Institute - DIBIT, Milan, Italy
Contributors

LINA MATERA • Laboratory of Tumor Immunology, Department of Internal Medicine, University of Turin, Italy
MOHAMAD MOHTY • Laboratoire d’Immunologie des Tumeurs, Institut Paoli-Calmettes, Université de la Méditerranée, Marseille, France
SOLVEIG H. MORE • Division of Medical Microbiology and Immunology, Bernhard-Nocht-Institute for Tropical Medicine, Hamburg, Germany
EVELIEN NAESSENS • Department of Clinical Chemistry, Microbiology and Immunology, Ghent University, Belgium
TIM NIEHUES • Hematology and Oncology, University Hospital, Heinrich-Heine-University, Düsseldorf, Germany
CHARLOTTE M. NIEMEYER • Pediatric Hematology and Oncology, Department of Pediatrics, University of Freiburg, Germany
MICHAEL I. NISHIMURA • Department of Surgery, University of Chicago, IL
DANIEL OLIVE • Laboratoire d’Immunologie des Tumeurs, Institut Paoli-Calmettes, Université de la Méditerranée, Institut National de la Santé et de la Recherche Médicale U119, Marseille, France
GLYN D. PALMER • Center for Molecular Orthopaedics, Harvard Medical School, Boston, MA
CIRIACO A. PICCIRILLO • Laboratory of Immunology, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, MD
STEFANO PLUCHINO • Neuroimmunology Unit, Department of Neuroscience, San Raffaele Scientific Institute - DIBIT, Milan, Italy
GÉRALD J. PRUD’HOMME • Department of Pathology, McGill University, Montreal, Canada
SERGIO QUEROL • Barcelona Cord Blood Bank, Centre de Transfusió i Banc de Teixits, Hospital Durán i Reynals, Barcelona, Spain
NIKOLAI G. RAINOV • Walton Centre for Neurology and Neurosurgery, University of Liverpool, UK
MICHELE ROSENZWAJG • Laboratoire d’Immunologie, Institut National de la Santé et de la Recherche Médicale, Paris, France
TOHRU SAKAI • Department of Nutrition, University of Tokushima School of Medicine, Tokushima, Japan
JOSÉ CARLOS SEGOVIA • Hematopoiesis Project, CIEMAT, Madrid, Spain
JOOST H. N. SCHUITENMAKER • Department of Cell Biology and Histology, Academic Medical Center, University of Amsterdam, Netherlands
CHRISTIAN SCHULTZ • Department of Paediatrics, Medical University Lübeck, Germany
VOLKER SCHUSTER • Division of Pediatric Immunology, Department of Pediatrics, University of Leipzig, Germany
Contributors

MICHAEL R. SHURIN • Clinical Immunopathology, University of Pittsburgh Medical Center, Cancer Institute, Pittsburgh, PA
HAUKE SIEVERTS • Department of Hematology/Oncology, University Children’s Hospital of Heidelberg, Heidelberg, Germany
VERONIQUE STOVE • Department of Clinical Chemistry, Microbiology and Immunology, Ghent University, Belgium
INGOLF SUREL • Institute of Medical Immunology, Charité, Humboldt-University, Berlin, Germany
KOSTAS TRIANTAPHYLLOPOULOS • Bone and Joint Research Unit, William Harvey Research Institute, Barts and Royal London Medical School, Queen Mary, University of London, UK
KOICHIRO TSUI • Division of Cellular Therapy, Advanced Clinical Research Center, Institute of Medical Science, University of Tokyo, Japan
TAKAHIRO UEDA • Division of Cellular Therapy, Advanced Clinical Research Center, Institute of Medical Science, University of Tokyo, Japan
BRUNO VERHASSELT • Department of Clinical Chemistry, Microbiology and Immunology, Ghent University, Belgium
PEDRO VIEIRA • Department of Cell Biology and Histology, Academic Medical Center, University of Amsterdam, Netherlands
HANS-DIETER VOLK • Institute of Medical Immunology, Charité, Humboldt-University, Berlin, Germany
ARNE von BONIN • Division of Medical Microbiology and Immunology, Bernhard-Nocht-Institute for Tropical Medicine, Hamburg, Germany
THOMAS VRAETZ • Pediatric Hematology and Oncology, Department of Pediatrics, University of Freiburg, Germany
VOLKER WAHN • Children’s Clinic, Uckermark Hospital, Schwedt/Oder, Germany
HENNING WALCZAK • Division of Apoptosis Regulation, German Cancer Research Center, Heidelberg, Germany
KERSTIN WILLMANN • BD Biosciences, San Jose, CA
JOSH WU • Department of Dermatology, University of Colorado Health Science Center, Denver, CO
JIM XIANG • Saskatoon Cancer Center, Departments of Microbiology and Oncology, University of Saskatchewan, Saskatoon, Canada
M. RITA I. YOUNG • Research Service, Hines Veterans Affairs Hospital, Hines, IL, and Departments of Pathology and Otolaryngology, Loyola University Stritch School of Medicine, Maywood, IL
MARTIN ZENKE • Max-Delbrück-Center for Molecular Medicine Berlin, Germany
ANDREE ZIBERT • Department of Pediatric Hematology and Oncology, University Hospital, Heinrich-Heine-University, Düsseldorf, Germany
INTRODUCTION
Primary Immunodeficiencies Caused by Defects of Cytokines and Cytokine Receptors

Volker Wahn

1. Introduction

The immune system responds to antigenic stimulation with a complex array of molecular events involving antigen-presenting cells, B-cells, T-cells, and phagocytes. Cytokines and their respective receptors are intimately involved in regulating such immune responses. Their pivotal role can be illustrated in animal models for which certain cytokines or their receptors have been deleted.

It is beyond the scope of this review to discuss our knowledge on growth factor or receptor deficiencies derived from animal models. I would rather like to focus on observations in children with selective molecular defects. For example, pulmonary alveolar proteinosis in some patients has been found to be associated with mutations in the genes for granulocyte macrophage–colony-stimulating factor (GM-CSF) receptor β-chain, which is shared with receptors for interleukin (IL)-3 and IL-5. Certainly, in the future we should be aware of further pathogenic mutations in humans that, to date, have been demonstrated in experimental animals only.

The majority of mutations in cytokine or cytokine receptor genes result in inherited immunodeficiencies. Our current knowledge on such disorders, therefore, will be summarized.

2. Interleukin-1

There is one report on defective IL-1 production associated with immunodeficiency with some evidence of familiality (1). The siblings described...