M. Eric Gershwin
John M. Vierling
Michael P. Manns
Editors

Liver Immunology

Principles and Practice

Second Edition

Springer

Liver Immunology

M. Eric Gershwin • John M. Vierling
Michael P. Manns
Editors

Liver Immunology

Principles and Practice

Second Edition

Springer

Editors
M. Eric Gershwin
Division of Rheumatology,
Allergy and Clinical Immunology
University of California School of Medicine
Davis, CA, USA

Michael P. Manns
Department of Gastroenterology, Hepatology
and Endocrinology
Medical School of Hannover
Hannover, Germany

John M. Vierling
Medicine and Surgery
Baylor College of Medicine
Houston, TX, USA

ISBN 978-3-319-02095-2 ISBN 978-3-319-02096-9 (eBook)
DOI 10.1007/978-3-319-02096-9
Springer New York Heidelberg Dordrecht London

Library of Congress Control Number: 2013952928

Printed on acid-free paper

Springer is part of Springer Science+Business Media (www.springer.com)

The editors and authors of this book dedicate the text and its contents to Linie Moore, symbolic of her courage, dedication, imagination, and enthusiasm. Linie is one of a kind and is a true star in the struggle to find a cure for autoimmune disease.

"Gutta cavat lapidem"

Foreword

Meeting the Challenge of Patients with Immune-Mediated Liver Disease

Medicine, health, and health care are in a period of rapid evolution. As expenses continue to soar, societal pressure will push us to provide better health for less money to more people. These lofty aims are achievable, although not likely with our present delivery system.

New systems will require even more data-driven decisions and a more tailored, personalized approach to individuals. This personalized approach will in part depend on understanding data available in the individual genetic code which can help direct risk assessment as well as help us make better diagnostic and treatment decisions.

Our new, data-driven approach will be based on a better understanding of underlying mechanisms, with an increased focus on more directed or targeted therapy. This new edition of this textbook will help set the stage for those exciting changes.

Advances in liver disease include the emergence of imaging modalities to assess liver fibrosis (magnetic resonance elastography and ultrasound elastography) improved imaging of liver masses and noninvasive methods to assess the biliary tree and vascular structures. Immunohistochemical staining allows differentiation of tumors and infections as well as characteristics of some immune-mediated diseases.

The understanding of the genetic code has led to genome-wide association studies which hold the promise of new insights into potential pathogenetic pathways that can be then explored with more directed, functional studies. These associations are already being used to target therapies designed to ameliorate the inflammatory response. In cancer therapy, whole genome sequencing of tumors has now allowed specifically targeted therapy which may yield greater efficacy with fewer adverse side effects. Genetic polymorphisms have also been used to predict treatment responses and predict development of steatosis, among other uses.

Therapy for liver disease has emerged quickly in the past decade. Hepatitis B is suppressed long term with excellent clinical results while the vast majority of patients with Hepatitis C may soon be curable. Primary biliary cirrhosis is now treatable with ursodeoxycholic acid while primary sclerosing cholangitis is still lacking the effective therapy.

Our understanding of the basic and clinical aspects of immune-mediated liver disease is rapidly progressing and an excellent update, such as provided in the second edition of this classic textbook, is timely. There has been increased attention on immune diseases of the liver because of a rising increase in prevalence in some cases, in others because of new discoveries that may give us clues to pathogenesis, while in other instances improved management has been established. Several diseases among this collection of illnesses remain without effective therapy, spurring further research to find sorely needed treatment options. Furthermore, several of these diseases are accompanied by increased risk of malignancy, adding more to the urgency to better understand and treat these conditions.

This second edition begins with important information about the epidemiology and mortality of liver disease worldwide. These are followed by chapters related to basic immunology, application of liver immunology for diagnosis, and several excellent chapters that provide a

solid foundation for understanding immune-mediated liver disease including those associated with the biliary tree. A chapter on non-hepatic manifestations of immune-mediated liver disease helps provide context for how these diseases affect the patient overall.

There are chapters that discuss various discrete immunologically mediated infectious liver disorders including those related to bacteria, parasites, and all of the classic viruses. Chapters on the traditional autoimmune liver diseases; primary biliary cirrhosis, autoimmune hepatitis, primary sclerosing cholangitis as well as overlap syndrome are included. The breadth of this second edition is highlighted by chapters on alcoholic liver disease, nonalcoholic fatty liver disease, and drug-induced liver disease among others, which have potentially immunologic features, yet are not usually included among the immune-mediated liver diseases. More classic immune-mediated liver disease occurring in the setting of transplantation, whether graft vs. host disease or liver transplantation, are also included.

The edition ends with a forward-looking view of future directions and how we might meet the challenge of refractory patients, written by the editors jointly.

The editors of this second edition have consolidated an outstanding group of authors who are responsible for the various chapters. The book will serve as a comprehensive textbook for many liver diseases, especially those that have an immune-related pathogenesis. The text does not cover malignant, vascular, congenital, or cystic diseases. It leverages the focus on immune-mediated diseases to provide an in-depth and comprehensive overview of this important aspect of liver disease.

This book will serve as an excellent overview for this rapidly evolving field and should add to our understanding of the pathogenesis of these diseases, as well as provide insights that can be harnessed into helping improve the care of patients afflicted with these various immune-mediated diseases. This book will be valued by those learning about this field in training as well as by established experts in the field. The editors are to be congratulated for this important contribution.

Rochester, MN, USA Keith D. Lindor, MD

Preface

Recognition of the importance of the liver to health by Babylonians in the nineteenth century BCE stands in stark contrast to the relative obscurity of the liver in the minds of most educated adults today. Medical appreciation of the vital nature of the liver's diverse functions continues to evolve along with our efforts to better understand a multitude of hepatobiliary diseases caused by alcohol, xenobiotics, viruses, autoimmunity, and genetic diseases. The unanticipated success of liver transplantation in the absence of histocompatibility matching between donor and recipient showed that the hepatic environment is immunosuppressive. Further studies proved that liver transplantation also protected other transplanted organs from being rejected, indicating that the liver is truly an immunologic organ. Recent data provide new insights into the physiological roles of hepatocytes, sinusoidal lining cells, activated macrophages (Kupffer cells), cholangiocytes and stellate cells, and their modulation of T cells, natural killer (NK) cells, and NKT cells. Concurrently, studies of the pathogenetic mechanisms involved in hepatobiliary diseases have provided unequivocal evidence that the pathogenesis of virtually all hepatobiliary diseases involves inflammation involving the innate and/or adaptive immune responses. Progress in our understanding of the liver as an immune organ and immunopathogenesis of diverse hepatobiliary diseases provides hope that this knowledge will rapidly be translated into more effective therapies in the near future. These factors were the impetus for the third edition of Liver Immunology: Principles and Practice, which is directed to clinicians, investigators, and students. The editors are indebted to all of the authors who have donated their talents, intellects, and expertise to provide "state-of-the-art" contributions. All of us hope that this book will provide new perspectives of hepatobiliary physiology and pathophysiology and stimulate creative approaches to accelerate the pace of research progress in the field. Time has validated our belief that continued studies of immunology of the liver will ultimately improve the care and the prognosis of patients afflicted with a diverse array of hepatobiliary diseases. The editors have many people to thank, not the least of which are the contributors, all of whom worked very hard to have their manuscripts delivered on time and in the style we requested. However, we especially want to thank Nikki Phipps and Kathy Wisdom, our assistants at UC Davis, who worked so hard to make this book a reality.

Davis, CA, USA — M. Eric Gershwin, MD, FACP
Houston, TX, USA — John M. Vierling, MD, FACP
Hannover, Germany — Michael P. Manns, MD

Contents

Contributors

Guruprasad P. Aithal NIHR Nottingham Digestive Diseases Biomedical Research Unit, Nottingham University Hospital NHS Trust and University of Nottingham, Nottingham, UK

Fernando Alvarez Division of Gastroenterology, Hepatology and Nutrition, CHU Sainte-Justine, Montreal, QC, Canada

Einar S. Björnsson Department of Internal Medicine, National University Hospital of Iceland, Reykjavik, Iceland

Kirsten Muri Boberg Section for Gastroenterology, Department of Transplantation Medicine, Oslo University Hospital, Rikshospitalet, Oslo, Norway

Institute of Clinical Medicine, University of Oslo, Oslo, Norway

Dimitrios P. Bogdanos Department of Medicine, School of Health Sciences, University of Thessaly, Biopolis, Larissa, Greece

Institute of Liver Studies, Transplantation Immunology and Mucosal Biology, King's College London School of Medicine at King's College Hospital, London, UK

Cellular Immunotherapy and Molecular Immunodiagnostics, Institute of Research and Technology Thessaly, Larissa, Greece

Albert Bosch Laboratory of Enteric Virus, Department of Microbiology, Institute of Food Safety and Nutrition, University of Barcelona, Barcelona, Spain

Christopher L. Bowlus Division of Gastroenterology and Hepatology, University of California at Davis Medical Center, Sacramento, CA, USA

Angela Ceribelli Division of Rheumatology and Clinical Immunology, Humanitas Clinical and Research Center, Milan, Italy

Department of BIOMETRA, University of Milan, Milan, Italy

Roger W. Chapman Gastroenterology Unit, John Radcliffe Hospital, Oxford, UK

Cecilia B. Chighizola Department of Clinical Sciences and Community Health, University of Milan, Milan, Italy

Division of Rheumatology, Istituto G. Pini, Milan, Italy

Derek Doherty School of Medicine, Trinity College, Dublin, Ireland

Scott L. Friedman Icahn School of Medicine at Mount Sinai, New York, NY, USA

Bin Gao Laboratory of Liver Diseases, National Institute on Alcohol Abuse and Alcoholism, National Institutes of Health, Bethesda, MD, USA

M. Eric Gershwin Division of Rheumatology, Allergy and Clinical Immunology, University of California at Davis School of Medicine, Davis, CA, USA

Stevan A. Gonzalez Division of Hepatology, Annette C. and Harold C. Simmons Transplant Institute, Baylor All Saints Medical Center, Fort Worth, TX, USA

Jan Grabowski Helmholtz Centre for Infection Research, Braunschweig, Germany

Emina Halilbasic Division of Gastroenterology and Hepatology, Department of Internal Medicine III, Medical University of Vienna, Vienna, Austria

Mathias Heikenwalder Institute of Virology, Technische Universitat Munchen/Helmholtz Zentrum Munchen, Munich, Germany

Gideon M. Hirschfield Centre for Liver Research, Institute of Biomedical Research, NIHR Biomedical Research Unit, University of Birmingham, Birmingham, UK

Harald Hofer Division of Gastroenterology and Hepatology, Department of Internal Medicine III, Medical University of Vienna, Vienna, Austria

Yaron Ilan Gastroenterology and Liver Units, Department of Medicine, Hadassah Hebrew University Medical Center, Jerusalem, Israel

Pietro Invernizzi Division of Rheumatology, Allergy and Clinical Immunology, University of California at Davis School of Medicine, Davis, CA, USA

Gerardo Kaplan Laboratory of Emerging Pathogens, Center for Biologics Evaluation and Research, Food and Drug Administration, Bethesda, MD, USA

Percy Knolle Institute of Molecular Immunology, Klinikum rechts der Isar, Technische Universitat Munchen, Munchen, Germany

Institutes of Molecular Medicine and Experimental Immunology, University Hospital, Friedrich-Wilhelms-Universitat Bonn, Munchen, Germany

Alexander Koch Department of Gastroenterology, Hepatology and Intensive Care Medicine, University Hospital Aachen, Aachen, Germany

Gadi Lalazar Department of Medicine, Marion Bessin Liver Research Center, Albert Einstein College of Medicine of Yeshiva University, Bronx, NY, USA

Patrick S.C. Leung Division of Rheumatology, Allergy and Clinical Immunology, University of California at Davis School of Medicine, Davis, CA, USA

Ka-Kit Li Centre for Liver Research and NIHR Biomedical Research Unit, University of Birmingham, Birmingham, UK

Liver Unit, Queen Elizabeth Hospital, Birmingham, UK

Zhe-Xiong Lian Department of Microbiology and Immunology, School of Life Sciences, University of Science and Technology of China, Hefei, Anhui, China

Rodrigo Liberal Institute of Liver Studies, King's College London School of Medicine at King's College Hospital, London, UK

Faculty of Medicine, University of Porto, Porto, Portugal

Keith D. Lindor Division of Gastroenterology and Hepatology, Mayo Clinic Foundation, Rochester, MN, USA

Sebastian Lunemann Department of Gastroenterology, Hepatology and Endocrinology, Hannover Medical School, Hannover, Germany

Xiong Ma Division of Gastroenterology and Hepatology, Ren Ji Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai Institute of Digestive Disease, Shanghai, China

Key Laboratory of Gastroenterology and Hepatology, Ministry of Health (Shanghai Jiao-Tong University), Shanghai, China

Ian R. Mackay Department of Biochemistry & Molecular Biology, Monash University, Clayton, VIC, Australia

Said Al Mamari Transitional Gastroenterology Unit, Oxford University Hospitals, Oxford, UK

Manisha Mandal Department of Physiology and Biophysics, KPC Medical College Hospital, Kolkata, West Bengal, India

Shyamapada Mandal Laboratory of Microbiology and Experimental Medicine, Department of Zoology, University of Gour Banga, Malda, West Bengal, India

Michael P. Manns Department of Gastroenterology, Hepatology and Endocrinology, Medical School of Hannover, Hannover, Germany

Wajahat Z. Mehal Section of Digestive Diseases, Yale University, New Haven, CT, USA

Pier Luigi Meroni Department of Clinical Sciences and Community Health, University of Milan, Milan, Italy

Division of Rheumatology, Istituto G. Pini, Milan, Italy

Giorgina Mieli-Vergani Paediatric Liver, GI & Nutrition Centre and Institute of Liver Studies, King's College London School of Medicine at King's College Hospital, London, UK

Institute of Liver Studies and Paediatric Liver, GI & Nutrition Centre, King's College London School of Medicine at King's College Hospital, Denmark Hill, London, UK

Mario U. Mondelli Department of Infectious Diseases, Fondazione IRCCS San Matteo, Pavia, Italy

Department of Internal Medicine, University of Pavia, Pavia, Italy

Francesco Negro Divisions of Gastroenterology and Hepatology and of Clinical Pathology, University Hospital, Geneva, Switzerland

James Neuberger Centre for Liver Research and NIHR Biomedical Research Unit, University of Birmingham, Birmingham, UK

Liver Unit, Queen Elizabeth Hospital, Birmingham, UK

National Health Service Blood and Transplant, Bristol, UK

Cliona O'Farrelly Comparative Immunology Group, School of Biochemistry and Immunology, Trinity College, Dublin, Ireland

Timoklia Orfanidou Cellular Immunotherapy and Molecular Immunodiagnostics, Institute of Research and Technology Thessaly, Larissa, Greece

Vaishali Patel Department of Gastroenterology, Hepatology & Nutrition, Virginia Commonwealth University, Richmond, VA, USA

Center for Clinical and Translational Research, Richmond, VA, USA

Robert P. Perrillo Division of Hepatology, Annette C. and Harold C. Simmons Transplant Institute, Baylor University Medical Center, Dallas, TX, USA

Rosa M. Pintó Laboratory of Enteric Virus, Department of Microbiology, Institute of Food Safety and Nutrition, University of Barcelona, Barcelona, Spain

Ulrike Protzer Institute of Virology, Technische Universitat Munchen/Helmholtz Zentrum Munchen, Munich, Germany

Ali Raza Transplant Hepatology Fellow, Baylor College of Medicine, St. Luke's Episcopal Hospital, Houston, TX, USA

Eirini I. Rigopoulou Department of Medicine, School of Health Sciences, University of Thessaly, Biopolis, Larissa, Greece

Maria De Santis Division of Rheumatology and Clinical Immunology, Humanitas Clinical and Research Center, Milan, Italy

Department of BIOMETRA, University of Milan, Milan, Italy

Arun J. Sanyal Department of Gastroenterology, Hepatology & Nutrition, Virginia Commonwealth University, Richmond, VA, USA

Center for Clinical and Translational Research,Richmond, VA, USA

Carlo Selmi Division of Rheumatology and Clinical Immunology, Humanitas Clinical and Research Center, Rozzano, Milan, Italy

Department of BIOMETRA, University of Milan, Milan, Italy

Division of Rheumatology, Allergy and Clinical Immunology, University of California at Davis, Davis, CA, USA

Daniel S. Smyk Institute of Liver Studies, Transplantation Immunology and Mucosal Biology, King's College London School of Medicine at King's College Hospital, London, UK

Katharina Staufer Division of Gastroenterology and Hepatology, Department of Internal Medicine III, Medical University of Vienna, Vienna, Austria

Christian P. Strassburg Department of Medicine I, University of Bonn Medical Center, Bonn, Germany

Gyongyi Szabo Department of Medicine, University of Massachusetts Medical School, Worcester, MA, USA

Hiroki Takahashi Department of Gastroenterology and Hepatology, JIKEI University Graduate School of Medicine, Tokyo, Japan

Zhigang Tian School of Pharmaceutical Sciences, Institute of Immunopharmacology and Immunotherapy, Shandong University, Jinan, Shandong, China

Department of Microbiology and Immunology, School of Life Sciences, University of Science and Technology of China, Hefei, Anhui, China

Michael Trauner Division of Gastroenterology and Hepatology, Department of Internal Medicine III, Medical University of Vienna, Vienna, Austria

Christian Trautwein Department of Gastroenterology, Hepatology and Intensive Care Medicine, University Hospital Aachen, Aachen, Germany

Palak Jitendrakumar Trivedi Centre for Liver Research and NIHR Biomedical Research Unit, University of Birmingham, Birmingham, UK

Liver Unit, Queen Elizabeth Hospital, Birmingham, UK

Stefania Varchetta Department of Infectious Diseases, Fondazione IRCCS San Matteo, Pavia, Italy

Diego Vergani Paediatric Liver, GI & Nutrition Centre and Institute of Liver Studies, King's College London School of Medicine at King's College Hospital, London, UK

John M. Vierling Division of Abdominal Transplantation, Baylor College of Medicine, St. Luke's Episcopal Hospital, Houston, TX, USA

Heiner Wedemeyer Department of Gastroenterology, Hepatology and Endocrinology, Hannover Medical School, Hannover, Germany

Mikio Zeniya Department of Gastroenterology and Hepatology, JIKEI University Graduate School of Medicine, Tokyo, Japan

Cai Zhang School of Pharmaceutical Sciences, Institute of Immunopharmacology and Immunotherapy, Shandong University, Jinan, Shandong, China

Renqian Zhong Shanghai Changzheng Hospital, Second Military Medical University, Shanghai, China

Introduction

1

Ian R. Mackay

The Arousal of Immunology

The invitation to prepare a foreword to "Liver Immunology Second Edition" is indeed an honor, given the successes of the first two editions of the book. The young and exuberant science of immunology, having just passed its first centennial and undergone detachment from microbiology in the 1950s, has enjoyed accelerating progress, at the laboratory and medical levels. Selection of the most influential advances in immunology in the "modern" era (post-1940s) is of course subjective, but would span topics as diverse as self-recognition to the immunological role of the intestinal microbiota. My list of the more significant advances is shown in Table 1.1.

The title of this text, *Liver Immunology*, subsumes notions of a "lymphoid liver" [1] and the liver as a "unique immunological organ" (Chap. 4) and thus a legitimate constituent among tissues intrinsic to the immune system. The liver sturdily fulfils essential immune defensive needs as a "gatekeeper" located strategically between the intestinal/portal and systemic blood circulations. Therefore, it can intercept influxes via the portal vein of microbial escapees or incompletely digested dietary constituents. This role depends on the liver being well equipped within its fenestrated vascular sinusoids, with all cellular elements of innate immunity, macrophages, myeloid dendritic cells, Kuppfer cells and mucosal-associated invariant T (MAIT) cells [2], with all these complemented by barrier functions of liver sinusoidal epithelial cells. Notwithstanding the tolerogenic capabilities of these sinusoidal cellular elements, our "lymphoid liver" can, and does, succumb to diseases due to dysfunction of its protective immune armory, whether as a result of ineffectual responses to hepatotropic viruses, or loss of self-tolerance with troublesome autoimmunity affecting hepatocytes or terminal cholangioles, or adverse reactivities to drugs disposed of by the intra-hepatic cytochrome P450 (CYP450) family of oxidative enzymes. Each of these dysfunctions can induce ongoing destructive inflammation—and it doesn't end there! Thus, various "degenerative" liver diseases exist, some common and others rare, in which endogenous products of hepatocellular injury resulting from ethanol abuse, metabolic steatosis, genetic deficiencies of the serpin (serum protease inhibitor) *alpha*-1 anti-trypsin provoke a cytokine-dependent auto-inflammatory response by defensive cells of the innate immune system. These issues are so expertly covered in the Chapters herein to follow that this Foreword could well conclude simply by commending the Editors on their judicious selection of contributing authors. However, I will use the Foreword as a rationale for the notion of "liver immunology" and explore some of the refractory questions that continue to challenge us.

Virus-Induced Chronic Inflammatory Hepatitis

In times long past, the only recognized type of persisting hepatitis was that known as "chronic active hepatitis" for which an autoimmune basis was eventually proposed. The very different situation today is that chronic viral hepatitis has become the overwhelmingly prevalent type, attributable to changed social customs and lifestyles, and readily available sensitive laboratory tests for the now identified causative viruses. Although many viruses have hepatotropic potential, it is only hepatitis viruses B and C (HBV, HCV) that do establish a chronic infection due to a non-eliminative host immune response to persisting intracellular virus provoking inflammation, recurring liver cell necrosis, regeneration, and fibrosis, culminating in cirrhosis and, sometimes, hepatocellular carcinoma. HBV and HCV infection of the liver in some respects are similar, but differ substantially including their capacity to establish a persistent infection.

I.R. Mackay (✉)
Department of Biochemistry & Molecular Biology,
Monash University, Clayton, VIC 3800, Australia
e-mail: ian.mackay@monash.edu

M.E. Gershwin et al. (eds.), *Liver Immunology: Principles and Practice*,
DOI 10.1007/978-3-319-02096-9_1,

Table 1.1 Some of the major discoveries in immunology 1940–2000

Timeline	Discovery
1945	Demonstration of specific cell (lymphocyte)-based immune reactivity in contrast to serum-based reactivity
1948	Immune tolerance as basis of self-non-self discrimination
1957	Clonal selection theory of acquired immunity
1960s	Failure of tolerance and ensuing autoimmunity as a cause of many diseases
1961–1963	Thymus as the site for progenitors of lymphocytes and tolerogenesis
1960s	Increasing interest in genetic determinants of immunological expressions—in mice (beginning 1940s) and in humans (beginning 1950s)
1969	The T and B lymphocyte paradigm
1970s	"Molecularization" of immunology—massively influential—e.g., multiple gene recombinations explained diversity of B and T cell antigen receptors
1970s	New T-cell subset (suppressor/regulatory T cells) down-regulates immune reactivities and explains peripheral immune tolerance—homeostasis
1980s	Identification in tissue extracts of "factors" antecedent to characterized cytokines, receptors, and intracellular signaling in immune responses
1990s	Essential inter-dependence of innate and adaptive immune systems broader visions of "innate" immunity
1990s	Recognition of importance of apoptosis and mechanisms as a component of immunological function
2000s	Role of intestinal commensal microbiota in shaping physiological and pathological immune responses

Chronic hepatitis B Carrier rates of HBV globally differ depending on socio-cultural lifestyles, routes of viral transmission, and racial-genetic background. In high prevalence regions transmission can be by vertical infection mother to fetus, or by close perinatal contact, whereas in low prevalence regions transmission is mostly parenteral, often in the setting of intravenous drug use. The vigor of adaptive immunity in healthy individuals ensures clearance of infection in some 95 % of instances, while failure to clear infection depends on immunodeficiencies associated with developmental immaturity, nutritional deficit, or coexisting chronic illness, e.g., renal failure. However, we need far more knowledge on the nature of the permissive immune deficiency states that underlie susceptibility such as general debility, malnutrition associated with poverty, alcohol or drug abuse, or chronic illness. A decreased T cell responsiveness to HBV appears important, perhaps by limiting the capacity for engagement of multiple antigenic epitopes presented by the virus. However, even so, among chronically infected individuals, there is a 2 % per annum viral clearance rate associated with demonstrable HBV-specific T cells and anti-HBs in blood [3].

Interestingly, with failure of viral clearance, a default option for the host is tolerogenesis resulting in a "healthy carrier" state. Immunogenetic factors have some influence on the occurrence or outcome of infection with HBV, and also the response to the normally highly effective HBV vaccine, noting that different HLA alleles appear to provide protection or susceptibility among different populations [4], and there are small effects of polymorphisms of the promoter for cytokine genes, IL-10 and TNF-alpha [5].

Since direct correlation has been drawn between viral load and propensity to progress to cirrhosis [6], therapeutic reduction of viral load is beneficial. However, the relative participation of CD4 and CD8 T cells in hepatocyte injury requires more study, while B cells enter the picture due to ongoing antigenic stimulation by viral antigens, leading to immune complex disease and/or essential mixed cryoglobulinemia [7].

Chronic hepatitis C HCV is less complex genetically and structurally than HBV, but equally illustrates the complexity of immune interactions between a "survival-adapted" virus and its human host [8]. In healthy individuals acute infection with HCV versus HBV is less readily cleared (~30 % versus ~95 %); and although debility-related immune deficiency predisposes, it is not necessary for chronic HCV infection. Nor is there a tolerance option as with HBV infection, since most if not all HCV carriers have some degree of hepatic inflammation. HCV hepatitis seems to be facilitated by various comorbidities, and particularly by effects of alcoholic or nonalcoholic steatosis, noting the propensity of HCV itself to induce fat deposition in liver cells [9]. Innate immunity provides a first line of defense against HCV infection, since Toll-like receptors (TLR) on phagocytic cells recognize pathogen (virus)-associated molecular patterns (PAMP). The RNA of HCV engages TLR3, thereby activating signaling pathways for expression of pro-inflammatory and anti-viral cytokines, particularly interferons (IFNs), and primes the host for an adaptive immune response [8]. While ensuing IFN expression results in some reduction in levels of HCV in liver cells, full clearance requires additionally a rapid and effective adaptive immune response involving engagement by T cells and likely B cells of multiple antigenic epitopes of the virus polyprotein [9], particularly by the NS5A protein of the virus [10]. There has been good progress in defining epitopes on structural and nonstructural proteins of the HCV particle, their relative capability of being presented by different HLA molecules, and activation of protective CD4 and CD8 T cell immunity. Comparable to HBV infection, the outcome of an acute infection depends on the quality and number of HCV epitopes initially engaged and efficient development of effector/memory T cells [11].

The many explanations for the escape of HCV from the host immune attack include ongoing development of

immunologically variant quasispecies that outrun the available T cell specificities of the host, suppression of T cell activities by HCV proteins, tardiness of primed T cells to move rapidly to the newly infected liver, defective engagement of critical HCV epitopes such as NS5A that favors viral persistence by exerting anti-apoptosis effects on infected hepatocytes [10], depletion of CD8 T-cell responsiveness during evolution of infection [11], and debility-related immune impairment of T cell and NK cell performance, with limited IFN-gamma responses. Finally the first encounter between naïve T cells and HCV occurs in the tolerogenic milieu of the liver rather than in the immunogenic milieu of a regional lymph node [12]. Among genetic influences, HLA class I and class II alleles influence clearance, well illustrated for the highly protective class I allele HLA B27 that engages an epitope within the NS5B protein, although structural polymorphisms of HCV evolve to circumvent this [13].

Initially in the chronic liver-damaging phase of HCV infection CD4 helper and CD8 cytolytic T cells (CTLs) are operative with good control of viremia albeit associated with greater evidence of histologic liver damage [14], whereas later T cell activity wanes but, even then, CTL activity is still demonstrable among T cells in liver, although not in blood. B cells have received relatively less attention in the host interaction with HCV, although antibody to HCV is clearly demonstrable and is directed to multiple components of the HCV polyprotein. Anti-HCV antibody does have neutralizing capacity, at least in infected chimpanzees, and likely serves to limit cell-to-cell transfer of virus in the liver.

The B-cell response becomes relevant in the later pathology of HCV infection in dictating many of the extrahepatic features [15, 16], particularly type 2 mixed cryoglobulinemia prevalent in endemic regions of infection [17]. These cryoglobulins contain HCV, anti-HCV, and oligoclonal IgM rheumatoid factor and are pro-inflammatory, causing arthralgia, vasculitis, cutaneous purpura, and membranoproliferative glomerulonephritis. Production is antigen (HCV)-driven since therapeutic reduction of viral load is ameliorative [17]. Another B cell feature of chronic HCV infection is production of various nonorgan-specific autoantibodies (NOSA) at relatively low levels [18], as described below. Later in the course, B cells may undergo lymphoproliferative expansion towards B-cell lymphoma resulting from chronic antigen drive with lymphomagenic chromosomal translocations such as the apoptosis inhibitory gene BCl-2 from chromosome16 to the IgH locus on chromosome 14 [t(14;18)(q32;q21.3)]; however, the one study on human HCV-infected liver tissue did not confirm this [19].

Conventional treatment of chronic hepatitis C was initially with anti-viral type 1 IFN but in recent years "big pharma" has been developing anti-viral drugs of ever-increasing efficacy that, used in combinations, clearly contain or even eliminate HCV and thereby "cure" the disease. Finally those seeking clues to the causes in general of autoimmune disease find it intriguing that conventional therapy of HCV infection with type 1 IFN provokes autoimmune reactions *de novo* affecting the thyroid gland [20] andother tissues.

Autoimmune Chronic Inflammatory Liver Disease

Autoimmune hepatitis (*AIH*) Knowledge on AIH has accumulated to such an extent over the past 60 years that readers could be readily forgiven for believing that all that needs to be known is already known. Yet even after Liver Immunology 1e was published in 2003, more knowledge on AIH has accrued. Fortunately, hepatologists are beneficiaries of the wisdom of thought-leaders, the International Autoimmune Hepatitis Group (IAIHG) that evolved from a conference on AIH in 1993 and has convened regularly thereafter. The IAIHG works to rationalize and standardize nomenclature, develop criteria to assist clinical diagnosis and epidemiologic studies, adjudicate on therapies, and promote research into AIH in general [21, 22]. A recent simplification in 2008 limited the cumbersome initial diagnostic criteria to just these items: negative indices of hepatitis virus infection, hypergammaglobulinemia, compatible histologic features (interface lymphocytic hepatitis with prominent plasmacytosis), and autoantibodies at requisite levels to prescribed autoantigens [23]. These simplified criteria do perform well but validation is needed.

However, notwithstanding all the advances, several problematic aspects to AIH remain unsolved, as follows.

Hyper-immunoglobulinemia (*hyper-IgG*). Recognized from the1950s, the earliest days AIH [24], extreme polyclonal hyper-IgG associates with the activity of the disease. It even provides a useful marker of response to treatment and aligns well with the plasmacytosis in the liver (and bone marrow). The usual but not entirely convincing explanation is that this hyper-IgG is simply a polyclonal immune response to degraded liver cells. Forthcoming genetic studies may provide some answers.

AIH-associated autoantibodies. The traditional diagnostic autoantibodies were discovered in the 1960s using indirect immunofluorescence (IIF) on frozen tissues: (a) nuclear chromatin (antinuclear antibody, ANA), (b) smooth muscle in rodent gastric mucosa (SMA) with a later recognition that filamentous (F) actin was the likely reactive moiety specific for (AIH), and (c) microsomes (cytoplasm-derived elements) of liver and kidney tubular cells (LKM, later called LKM1). Subsequently a mutual exclusivity in reactivity of sera for ANA/SMA or anti-LKM led to the specification of two types, 1 and 2, of AIH (see below). These antibodies underpin

laboratory diagnosis, but, at the same time, exemplify a complex unsolved puzzle—why the association of a given autoimmune disease such as AIH with an autoantibody directed to a molecule that has no discernible correlation with the cellular pathology? Various other examples would include the diagnostic autoantibodies detected in Sjogrens disease, polymyositis, and systemic sclerosis.

Also recognized in AIH are other autoantigens of practical and/or theoretical interest [25]. These include a cytoplasmic constituent named "liver-pancreas-soluble liver antigen" (LP/SLA), identified as UGA-serine transfer (t)-RNA protein complex, to which antibody can be diagnostic in otherwise pan-seronegative cases and/or point to severe progressive disease. Another antigen of interest is the reactant for what was first called "granulocyte-specific ANA" [26], and later atypical anti-neutrophil cytoplasmic antibody (ANCA). This antibody is detectable at high prevalence in AIH but only in type 1 and not type 2. Thought has reverted to the reactant being an unidentified neutrophil nuclear rather than a cytoplasmic constituent. A further reactant of interest seen in type 2 AIH elicits an autoantibody that is a fellow-traveler with anti-LKM; this liver cytosol antigen (LC-1) has been molecularly identified as formiminotransferase cyclodeaminase. Since, occasionally, anti-LKM$^+$ ve sera react with CYP450 isoforms other than the prototypic 2D6 (often in drug-induced forms of hepatitis) and are directed against the P450 isoform, e.g., 2C9 or 3A1, that hydroxylates the drug, is there an as yet undetected molecule that, in the course of its disposal by CYP450 2D6, initiates the apparently spontaneous anti-LKM$^+$ ve AIH?

Specificity of anti-F actin for AIH. The designation "SMA" for one of the major serological reactants in AIH is so embedded that any change is unlikely, but my perception (and perhaps not all would agree) is that the true AIH-relevant reactant is filamentous (F) actin, whether detected by IIF testing by reactivity with actin microfilaments in renal glomeruli and tubules [25], or by ELISA with purified F actin, and that type 1 AIH is the single disease in which anti-F actin is regularly demonstrable. Moreover, positivity for anti-F actin helps to separate AIH from viral and other miscellaneous causes of low-level SMA reactions with other cellular filaments, and also from SLE with which AIH is occasionally aligned. F actin is relatively neglected as an autoantigenic molecule, since it has attracted little interest in its immunoreactivity or relationships between its epitope sites and the functional binding sites for some 70 cytoplasmic proteins among which is its cell motility partner, myosin.

Two serological types of AIH. The concept of two "serotypes" of AIH, 1 and 2, evolved from observations in 1987 on the mutual exclusivity in AIH of sero-positivity of ANA/SMA (type 1) and anti-LKM (type 2) [27]. The distinction is matched by differing HLA susceptibility alleles. Interestingly type 1 aligns more with multisystem nonorgan-specific diseases, whereas Type 2 more with organ-specific diseases. No convincing liver-specific autoantigen can be distinguished in type 1 AIH (despite much effort to identify a liver membrane—specific antigen), whereas the LKM1 reactant has been molecularly identified as the 2D6 isoform enzyme of CYP450 family allowing for development of useful mouse models which are lacking for the more prevalent type 1 AIH. Two different modes of immunopathogenesis for the one clinical disease in the one single organ are indeed very curious [28].

NOSA in chronic HCV infection. The possibility that the disease-defining AIH-associated autoantibodies could result entirely from a B-cell response to destruction and spillage of liver cell constituents seems untenable given the existence of the two disease serotypes, each with a distinct antibody profile. Yet issue injury in itself, e.g., ischemic infarction, is known to evoke a low-level immune response, expressed histologically by lymphocytic infiltration. This, then, likely explains the low levels of NOSA in chronic hepatitis C (CHC) as described in European studies such as that of Stroffolini et al. [19]; the prevalence in CHC of any NOSA reached approximately 36.9 %, and for ANA ~16 % and SMA ~27 %, although anti-LKM1 reached only ~2 %, similar to that (~5 %) among healthy hepatitis C virus carriers in similar locations. Disease if any in these HCV carriers is not typical of AIH. Also, and, in contrast to spontaneous type 2 AIH, the autoantibodies may react with CYP450 isoforms other than 2D6, or with epitopes on CYP450 2D6 other than those engaged by type 2 AIH sera. There being no association between any of these NOSA and hepatitis disease expressions, these pathogenetically irrelevant autoantibodies should sound a warning note to clinicians on over-interpretation of results of serological laboratory assays.

T cells in liver cell injury of AIH. In type 1 AIH T cells are prominent in the lymphocytic infiltrates in the liver and are presumed by some authors to determine liver cell damage. However, in type 1 AIH, relevant autoantigen preparations are not available, and hence assay systems for cytotoxic or cytokine-releasing T cells are inapplicable, as pertains for various of the multisystem autoimmune diseases. On the other hand, in sero-type 2 AIH, T cells in blood do respond to immunoreactive peptides derived from the characterized autoimmune reactant CYP450 2D6.

T reg cells and immunological homeostasis in AIH. In his classic monograph on clonal selection theory in 1959, FM Burnet developed the idea of forbidden (self-reactve) clones of lymphocytes and envisaged that healthy individuals must possess *homeostatic mechanisms* to render these ineffective. Now, over 50 years later, immunological homeostasis has

regained currency through the agency of regulatory T cells (Tregs) that in some way serve to nullify anti-self-reactivities in the periphery. The corollary is that defects in numbers or function of Tregs are complicit in autoimmunity. There are already hints of such processes in the pathogenesis of autoimmune liver diseases—presumably much more will be heard of Tregs in a range of autoimmune diseases.

Further details. The various issues concerning AIH engaged herein are examined in greater depth in Chaps. 7 and 19.

Primary biliary cirrhosis (*PBC*). I am rather familiar with the story of PBC. I still recall reading, even though over 50 years ago, the exemplary clinical research publication from the Rockefeller Institute that put PBC "on the map" [29], and wondering why so little was known of its cause. As fortune would have it, only several years later on we ascertained a positive result (albeit in just the one PBC case tested) with a complement fixation test for autoimmunity to a cellular cytoplasmic constituent—identified as mitochondria in London using IIF. This was telling us something! Detection of anti-mitochondrial antibody (AMA) became one of the most useful, and widely used, of all the immunoserologic diagnostic assays. But identification of the actual mitochondrial reactant progressed only slowly until the 1980s when we and others showed by immunoblot that this was a ~72 kDa polypeptide. Next a cDNA was isolated by molecular cloning from a gene expression library by one of the Editors of this text (MEG) working as a sabbatical visitor at the Hall Institute in Melbourne [30]. The elusive reactant for AMA, finally identified as the E2 subunit of enzymes of the 2-oxo-acid dehydrogenase complex (2-OADC) (chiefly pyruvate dehydrogenase), allowed for a stream of immunological studies including localization of immunodominant autoepitopes for antibody and T cells to the inner lipoyl domain of the E2 subunit (PDC-E2). This heralded novel insights into this enigmatic disease and posed the questions on why and how uncontrolled autoimmune responses to the autoepitope of PDE-E2 might occur, and how these might damage specifically the terminal cholangiolar cells, as seen in PBC.

Essentially the "core" autoepitope is a highly conserved linear sequence (residues 169–176, IETDKATIG) that includes lysine (^{173}K) to which is attached the lipoyl cofactor, although the "complete" antibody paratope might span residues within the conformational structure from ^{131}MH to F…V^{180}. Sooner or later, we may see a solved crystal structure of a monoclonal anti-PDC-E2 in a complex with purified PDC-E2. Studies on T-cells in PDC-E2 revealed reactivity to a similarly located epitope in the inner lipoyl region of PDC-E2 and, as expected, there was a very high enrichment, 150-fold, of PDC-E2 epitope-reactive CD4$^+$ T cells in liver infiltrates, and in portal lymph nodes compared with blood. Finally there is immunohistochemical evidence of invasion and destruction of biliary ductular cells by epitope-specific effector cytolytic CD8$^+$ T cells.

But there is an "elephant in the room!" That is, some 30–40 % of cases of PBC express another set of autoantibodies; these being to nuclear antigens. Why is this? These ANA, in contrast to those routinely studied in rheumatic diseases, are PBC-specific and show unique staining patterns by IIF. Mostly they are molecularly characterized. The specificities include (a) "speckled dot" representing the Sp100 molecule and related promonocytic leukemia (PML) protein, (b) "nuclear membrane" representing proteins gp210 and gp63 of the nuclear pore complex, and (c) centromeric protein (CENP) otherwise characteristic of limited cutaneous systemic sclerosis. These atypical ANAs provide no clues to provocative causes or pathogenesis of PBC and simply serve to place the disease in that "twilight zone" between Th1,Th17- dominant organ-specific and Th2-dominant multisystemic autoimmune diseases marked by deficient peripheral tolerance emanating from dysfunction of Treg cells.

The discovery of the molecular basis for AMA reactivity prompted a sustained research effort at Davis CA into all aspects of PBC, on the premise that unpicking the "genes and environment" nexus should prove fruitful. The results, compacted in Chap. 18, strengthen a belief that explanations for PBC, as for autoimmunity in general, will ultimately be resolved into effects of multiple possible genetic anomalies interactive in various ways with multiple possible environmental provocations—under conditions in which chance will have an influence of uncertain magnitude [31].

The genetic components in PBC might be seen as less prominent than those for other autoimmune diseases, yet there is a uniquely high concordance for PBC in monozygotic twins (~60 %); a strong intra-familial susceptibility, a notably high female predisposition, and data from a mouse model are supportive [32].

Coming to environmental components, attention has been directed to sources of epitope mimics of the PDC-E2 lipoyl domain autoantigen. These range from infections with microbes that carry versions of the 2-OADC enzymes to exposure to novel xenobiotics that structurally influence the PDC-E2 region so as to create an immunogenic mimic sufficiently resembling PDC-E2 to break tolerance to the natural epitope. But perhaps there is no need to invoke extrinsic agents as initiators of autoimmune disease given that products of defective (incomplete) apoptosis may serve this function, prompting use of the term "apoptope." Experimentally it was found that there are unique features to apoptosis of cholangiocytes in that these cells specifically lack the capacity for glutathionylation allowing PDC-E2 to remain intact in apoptotic blebs as a potential immunogenic apoptope [32]. Then, with tolerance broken, by whatever means, and forbidden clones established, PBC would become slowly established by ongoing reexposure of the lymphoid system to the natural autoantigen.

A still further outcome of the availability in PBC of molecularly characterized autoantigens has been the encouragement given to develop mouse models of the disease. Already, over the past decade, these have (a) indicated the likelihood that genetic influences are indeed important in pathogenesis [33]; (b) shown that environmental agents and particularly xenobiotics are candidate initiators of PBC [34]; and (c) revealed strong permissive influences exerted by defects in peripheral tolerance dependent on signaling pathways of the receptor for the polyfunctional cytokine transforming growth factor (TGF)-beta [35], with effector functions attributable to clonally restricted populations of autoantigen-specific $CD8^+$ ve T cells [36]. These mouse models illustrate that disrupted TGF signaling indeed influences immunological homeostasis—their disadvantage is the relatively short life span of the mouse, precluding close recapitulation of the slowly evolving human PBC.

Primary sclerosing cholangitis. There are unquestioned immunological accompaniments to this mysterious disease, with some suggestive of autoimmunity including a tendency to overlap with type 1 AIH, more particularly in childhood (Chap. 24), and a disease association with ulcerative colitis for which, however, an autoimmune basis is being questioned, and an association with the "autoimmune" HLA haplotype B8 DRB1*030I. However, there are too many incongruities for PSC in adults to be ascribed to autoimmunity: It is male-dominant, the cholangitic lesions are sparse in lymphocytes but rich in fibrocytes, no disease-specific marker autoantibody is demonstrable, although there is a high frequency (88 %) of an atypical pANCA (not of proteinase 3 specificity). As mentioned, children with type 1 AIH coexpress an associated cholangiolar disease called "autoimmune sclerosing cholangitis," but its relationship to adult PSC remains undefined: perhaps a preferable descriptor would be "pediatric autoimmune cholangitis."

So, in conclusion, we see adult PSC as an aberrant proinflammatory response to products of otherwise innocuous intestinal microorganisms with ensuing cytokine activation, and a periductular myofibroblast response—and thus essentially auto-inflammatory (see below).

Drug-Induced Chronic Inflammatory Liver Disease

Immune-mediated drug-induced liver injury (imDILI) (sometimes called "idiosyncratic") could depend on several mechanisms. One is conjugation of a reactive metabolite of the drug to a host protein which, in the case of liver, is likely to be the enzyme protein, e.g., a CYP450 isoform responsible for its disposal (Chap. 27). This adduct generates an antigenic moiety which, on a permissive genetic background, can promote the inductive phase of an immune response expressed as allergic sensitization to the drug, and with ensuing inflammatory reactivity by the host. The actual site of induction is uncertain, whether within the liver or regional (hilar) lymph nodes. The "executive" phase of the response is variable mechanistically, being either antibody or T cell dominant but, at present, neither in vitro nor in vivo test systems seem sufficiently well developed to define the process in each individual instance of imDILI.

Of note, imDILI is sometimes accompanied by production of autoantibodies that simulate those detected in spontaneous AIH, either of ANA/SMA positivity or of anti-LKM positivity. The former, which challenge explanation, were seen prototypically in hepatitis occurring after prolonged use of the now obsolete hypotensive drug, a-methyl dopa, and nowadays are seen (rarely) with use of minocycline, nitrofurantoin, flucloxacillin, and others. The latter anti-LKM type would intuitively be more frequent, since many drugs are enzymatically disposed of by hydroxylation by isoforms of the CYP450 family, with the antibody corresponding to the isoform that hydroxylates the drug. Examples include the uricosuric tienelic acid (no longer marketed) that is degraded by CYP450 2C9 and provoked imDILI accompanied by anti-LKM 2C9, and the anti-hypertensive hydrallazine that is degraded by CYP 1A2 and (infrequently) provokes imDILI accompanied by anti-LKM CYP 1A2. Perhaps the best diagnostic procedure is observing recovery from imDILI after identifying and ceasing therapy with the culprit drug and, if needed, a deliberate (and carefully supervised) rechallenge.

Alloimmune Chronic Inflammatory Liver Disease

Alloimmune liver disease occurs in the context of host-versus-graft (HVG) or graft-versus-host (GVH) reactions occurring after transplantation of a donor allogeneic liver, or after transplantation of donor allogeneic bone marrow (BM) cells reactive against host liver (and other tissues). The outcome is a potent immunologic response whether to the "foreign" major MHC (HLA) class 1 molecules (and likely class 2 as well) or, in the case of BM transplants from HLA-matched donors, with "minor" transplantation antigens. Reactivity is expressed as mixed combinations of inflammatory damage to hepatocytes (interface hepatitis), biliary cells with ductopenia, or blood vessels with vascular occlusive lesions. This physiological propensity for allo-antigenic reactivity is attenuated by the tolerogenic milieu of our lymphoid liver so that allogeneic liver or BM transplants tend to succeed well despite MHC barriers, compared with skin and kidney, so leading to liver and BM transplantation becoming thriving elements of applied immunology.

Liver allografts, *HVG hepatitis*. The relatively less aggressive responses by the host to liver allografts is exemplified in some species (pig and some rodent strain combinations) by success with no requirement for immunosuppression, and in humans a lesser than expected need for immunosuppressive drugs. Yet the liver cannot be regarded as "immunologically privileged" since it is freely accessed by the portal venous and arterial circulations. Finally, the inherent intra-hepatic tolerogenicity is claimed to be augmented by lymphocytic chimaerism due to leakage out from a grafted liver of donor leucocytes.

However, rejection reactions either acute or chronic against the liver allograft do occur in some 80 % of human liver allografts. Acute rejection is expressed as portal leucocytic (granulocyte, monocyte, lymphocyte) infiltration seen as interface hepatitis, biliary ductulitis with ductopenia, and vascular endothelitis. Chronic rejection is expressed particularly by biliary ductopenia and obliterative arteritis. It is intriguing that an AIH can develop in an allografted liver, expectedly if an autoimmune disease was the reason for the transplant, but surprisingly when it occurs "de novo" (as it usually does) in allografts done for diseases other than autoimmune liver disease [37]. The basis for this diagnosis is histology together with fulfillment of the other conventional AIH criteria. Indeed the occurrence of "de novo AIH" in a liver allograft pose intriguing questions for the genesis of autoimmune disease in general.

Hemopoietic cell allografts, *GVH hepatitis.* There are many applications of allogenic hemopoietic (bone marrow) cell transplantation (HCT) in contemporary practice including immunodeficiencies, hematologic malignancies, aplastic anemia and, increasingly, in treatment of intractably progressive autoimmune diseases.

GVH disease is a complication in some 30–50 % of cases of allogeneic HCT from HLA-matched siblings. It is attributable to mature T lymphocytes of the donor inoculum, having been "protected" by immunosuppression of the recipient, reacting with foreign (non-HLA) "minor" histocompatibility antigens of host origin that become exposd on the cell-surface. The tissues predominantly affected by GVH disease are skin, intestinal tract mucosal surfaces, and liver, particularly cholangiocytes. Expectedly, given the likely similarity of the mode of pathogenesis, expressions resemble those of multisystem autoimmune diseases. In the liver, as with HVG disease, the lesions are hepatitic resembling those AIH, cholangitic resembling those of PBC, or vascular and resemble (to a degree) those of primary systemic sclerosis (scleroderma). In particular, in both HVG and GVH disease, intrahepatic biliary ductular cholangiocytes appear highly vulnerable to immune attack [38], as pertains in PBC, and in both conditions there is destructive invasion of ductules by activated T lymphocytes. There was even a claimed detection of AMA in a mouse model of GVH disease with affected bile ducts, but AMA was scarce among the autoantibodies tested for among 95 cases of human GVH disease [39].

Congenital alloimmune hepatitis. This alloimmune gestational disease of the liver, previously described (erroneously) as neonatal hemochromatosis, can present as fetal death in utero or severe liver dysfunction in the neonate. Once established, the disease characteristically recurs in subsequent pregnancies. Whtington [40] proposed the likelihood of a maternal immune attack on a surface-exposed liver cell-specific alloantigen occurring mid-term in gestation, with production of IgG alloantibody that crosses the placenta and causes a complement-dependent lysis of fetal hepatocytes.

The question of course arises on the nature and identity of the provocative liver-specific alloantigen. One liver alloantigen well-known to us was first described in the 1960s in mice, as the liver-specific F antigen which is a highly conserved and abundant liver cytoplasmic protein among mammals, including humans, that carry one or other of the two allotypes, F-1 or F-2 [41]. Immunization with allotypic F breaks tolerance and raises precipitating antibody that reacts with both allelic forms, the non-self-immunogen and the self-protein, thus eliciting both allo- and autoantibody. Cloning of the genes encoding the murine F alloantigens revealed that the deduced protein products had 95 % homology with a notable sequence difference near the carboxy terminus [41], but provided no functional insights. The known properties of the F alloantigen give this protein a candidate status as the alloantigen that causes the congenital alloimmune hepatitis of human infants.

Auto-inflammatory Chronic Liver Diseases

"Autoinflammatory" is a term that is gaining currency to describe an inflammatory response *sui generis*. This response can be invoked particularly by cellular degradations and products thereof, likely involves processes of innate immunity including induction by cytokines and chemokines, and is independent of adaptive immune responses [42]. In the liver, these cellular degradations may be associated with products associated with lipid accumulations, protein misfolding diseases, heavy metals (iron, copper), or other cytoplasmic inclusions not adequately disposed of by chaperone pathways.

An exemplary "autoinflammatory" liver disease is alcoholic hepatitis in which lipid inclusions known as Mallory bodies excite a pericellular neutrophilic inflammatory reaction, T cell chemotaxis, pro-inflammatory Type 1 cytokine release, and progressive fibrosis (Chap. 22). Particular attention is now directed towards a newer entity which hepatologists became aware of in the early 1980s, styled nonalcoholic

steatohepatitis (NASH), or a wider category of nonalcoholic fatty liver disease (NAFLD) (Chap. 23). The "non-alcoholic" (NA) component of the acronym is a hangover from earlier days when fat in the liver was such a reliable marker of alcoholic liver damage abuse that any other cause could not be entertained. Although some writers have invoked an adaptive immune response to these lipid inclusions to explain progression of alcoholic hepatitis to cirrhosis, the data are tenuous and the more likely process is activation of the innate immune system with production by macrophages of pro-inflammatory chemokines and cytokines, in combination with other potentiating factors, as reviewed [42], and with a genetic component since in some cases steatosis can be quite innocuous.

The reason in the first place for fat accumulation in the liver in NASH is often (~85 % of cases) the multifactorial "metabolic syndrome," characterized by central obesity, hypertriglyceridemia, hypertension, Type 2 diabetes, and insulin resistance. There is evidence that secretion of leptin by adipocytes can contribute to attraction into adipose tissue of macrophages [43] which thereupon promote cytokine-driven pro-inflammatory responses mediated by TNF-alpha, and profibrogenic TGF beta. Thus, NAFLD (Chap. 23) that lies at an intersect of hepatology, metabolism, immunology, inflammation, and genetics has strong claims for inclusion in this contemporary text on liver immunology.

Tornada

Tornada is an Occitan literary term for a short piece at the end of a body of writing, often a poem. Here, my tornada is presented to conclude this condensed overview of the many emerging issues pertaining to immunohepatology and to commend "Liver Immunology Edition 3" enthusiastically as an authoritative and comprehensive conspectus of this burgeoning area of enquiry into liver function and pathology.

Acknowledgment My thanks to Ms Elaine Pearson for her kind help in preparation of the manuscript.

References

1. Kita H, Mackay IR, Van de Water J, Gershwin ME. The lymphoid liver: considerations on pathways to autoimmune injury. Gastroenterology. 2001;120:1485–501.
2. Kjer-Nielsen L, Patel O, Corbett A, et al. MR1 presents microbial vitamin B metabolites to MAIT cells. Nature. 2012;491:717–23.
3. Rehermann B, Naseimbeni M. Immunology of hepatitis B virus and hepatitis C virus infection. Nat Rev Immunol. 2005;5:215–29.
4. Godkin A, Davenport M, Hill AVS. Molecular analysis of HLA class II associations with hepatitis B virus clearance and vaccine non-responsiveness. Hepatology. 2005;41:1383–90.
5. Cheong JY, Cho SW, Hwant IL, et al. Association between chronic hepatitis B virus infection and interleukin-10, tumor necrosis factor-α gene promoter polymorphism. J Gastroenterol Hepatol. 2006;21:1163–9.
6. Iloeje UH, Yang H-I, Su J, et al. Predicting cirrhosis risk based on the level of circulating hepatitis B viral load. Gastroenterology. 2006;130:678–86.
7. Galli M, Careddu F, D'Armino A, Monti G, Messina K, Invernizzi F. Hepatitis B virus and essential mixed cryoglobulinaemia. Lancet. 1980;1:1093.
8. Gale M, Foy EM. Evasion of intracellular host defence by hepatitis C virus. Nature. 2005;436:939–45.
9. Perumalswami P, Kleiner DE, Lutchman G, et al. Steatosis and progression of fibrosis in untreated patients with chronic hepatitis C infection. Hepatology. 2006;43:780–7.
10. Szabo G. Hepatitis C, virus NS5A protein—a master regulator? Gastroenterology. 2006;130:995–9.
11. Bowen DG, Walker CM. Adaptive immune responses in acute and chronic hepatitis C virus infection. Nature. 2005;436:946–52.
12. Ward S, Lauer G, Walker B, Klenerman P. Cellular immune responses against hepatitis C virus: the evidence base 2002. Clin Exp Immunol. 2002;128:195–203.
13. Bowen DG, Zen M, Holz L, Davis T, McCaughan GW, Bertolino P. The site of primary T cell activation is a determinant of the balance between intrahepatic tolerance and immunity. J Clin Invest. 2004;114:701–12.
14. Neuman-Haefelin R, McKiernan S, Ward S, et al. Dominant influence of an HLA-B27 restricted CD8+ T cell response in mediating HCV clearance and evolution. Hepatology. 2006;43:563–72.
15. Nelson DR, Marousis CG, Davis GL, et al. The role of hepatitis C virus-specific cytotoxic T lymphocytes in chronic hepatitis C. J Immunol. 1997;158:1473–81.
16. Cacoub P, Poynard T, Ghillani P, et al. Extrahepatic manifestations of chronic hepatitis C. Arthritis Rheum. 2000;42:2204–12.
17. Pivetti S, Novarino A, Merico F, et al. High prevalence of autoimmune phenomena in hepatitis C virus antibody positive patients with lymphoproliferative and connective tissue disorders. Br J Haematol. 1996;95:204–11.
18. Agnello V, Abel G. Localization of hepatitis C virus in cutaneous vasculitic lesions in patients with type II cryoglobulinemia. Arthritis Rheum. 1997;40:2007–15.
19. Stroffolini T, Colloredo G, Gaeta GB, et al. Does an 'autoimmune' profile affect the clinical profile of chronic hepatitis C? An Italian multicentre survey. J Viral Hepa. 2004;11:1–6.
20. Sansonno D, Tucci FA, De Re V, et al. HCV-associated B cell clonalities in the liver do not carry the t(14;18) chromosomal translocation. Hepatology. 2005;42:1019–27.
21. Johnson PJ, McFarlane IG. Meeting report: International Autoimmune Hepatitis Group. Hepatology. 1993;18:998–1005.
22. Alvarez F, Berg PA, Bianchi FB, et al. International Autoimmune Hepatitis Group Report: review of criteria for diagnosis of autoimmune hepatitis. J Hepatol. 1999;31:929–38.
23. Hennes EM, Zeniya M, Czaja A, et al. Simplified criteria for the diagnosis of autoimmune hepatitis. Hepatology. 2008;48:169–76.
24. Mackay IR. Historical reflections on autoimmune hepatitis. World J Gastroenterol. 2008;14:3292–300.
25. Smalley MJ, Mackay IR, Whittingham S. Antinuclear factors and human leucocytes: reaction with granulocytes and lymphocytes. Australas Ann Med. 1968;17:28–32.
26. Mackay IR. A 50-year experience with autoimmune hepatitis: and where are we now. J Gastroenterol. 2011;46 Suppl 1:17–28.
27. Homberg J-C, Abuaf N, Bernard O, et al. Chronic active hepatitis with anti liver/kidney microsome antibody type 1: a second type of autoimmune hepatitis. Hepatology. 1987;7:1333–9.
28. Mackay IR. Autoimmune hepatitis: what must be said. Exp Mol Pathol. 2012;93:350–3.