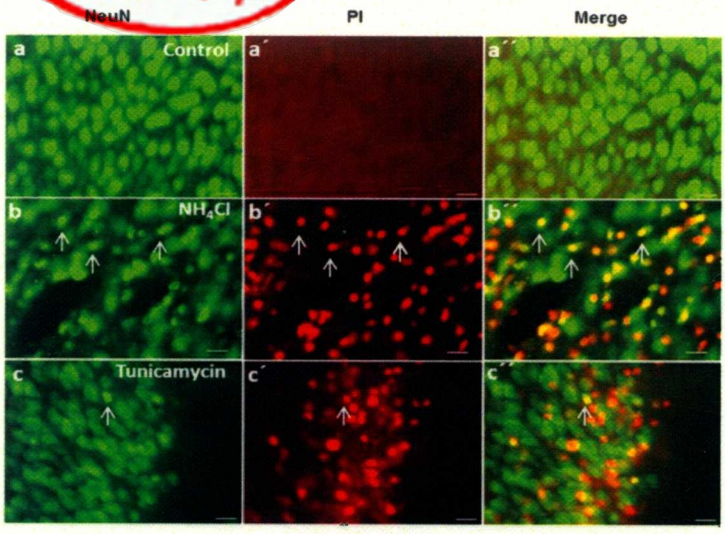
HEPATOLOGY RESEARCH AND CLINICAL DEVELOPMENTS

CHRONIC LIVER DISEASE

From Molecular Biology to Therapy



J. C. Perazzo * Francisco Eizayaga
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Alberto E. Muñoz * Néstor R. Lago
Editors

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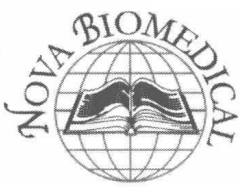

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LIST OF CONTRIBUTORS

Fernando J. Barreyro, MD, Ph.D
Laboratory of Microbiology, Faculty of Chemical and Natural Sciences,
National University of Misiones, Argentina.
Member of the National Council of Scientific and Technological Research (CONICET),
Argentina

Liliana Bianciotti, Biochem. Ph.D
Instituto de Inmunología, Genética y Metabolismo (INIGEM-CONICET-UBA),
Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires.
Professor of Pathophysiology, Faculty of Pharmacy and Biochemistry,
University of Buenos Aires, Buenos Aires, Argentina.

Yanina Bota
Instituto de Inmunología, Genética y Metabolismo (INIGEM-CONICET-UBA),
Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires.

Carlos Brodersen, MD, Ph.D
Chair of the Gastroentherology Unit Durand Hospital, Ciudad de Buenos Aires, Argentina.
Professor of Internal Medicine, University of Buenos Aires, Buenos Aires, Argentina.

Alicia Brusco, Biochem. PhD
Director of the Instituto de Biología Celular y Neurociencia
"Prof. E. De Robertis" (UBA-CONICET).
Facultad de Medicina, Universidad de Buenos Aires, Argentina

Laura Caltana, MD, PhD
Instituto de Biología Celular y Neurociencia "Prof. E. De Robertis" (UBA-CONICET).
Facultad de Medicina, Universidad de Buenos Aires, Argentina

Luis Cassinotti, Msc
Doctoral fellow at catedra de Fisiología-Instituto de Química y Metabolismo del Fármaco (IQUIMEFA-CONICET-UBA)
Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires

Sara Chao, MD
Gastroenterology Division in President Perón Hospital. Avellaneda, Buenos Aires, Argentina.
Intensive Care Specialist at the Critical Care Division, Marie Curie Hospital, Buenos Aires, Argentina.

Francisco Eizayaga, MD, Ph.D
Senior Researcher of the Laboratory of Hepatic Encephalopathy and Portal Hypertension
Department of Pathophysiology, University of Buenos Aires,
Ciudad de Buenos Aires, Argentina.
Professor, Maimonides University, Buenos Aires, Argentina.

Maria Gimena Fernandez, MD.
Hepatologist of the Churruca Hospital, Buenos Aires, Argentina

Bernd L. Fiebich, Ph.D
Department of Psychiatry. Director of the Neurochemistry Lab,
University of Freiburg Medical School, Freiburg, Germany

Diego Flichman, Biochem. Ph.D
Professor of Virology, Facultad de Farmacia y Bioquímica,
Universidad de Buenos Aires, Buenos Aires, Argentina.
Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET)

María Julia Guil, Biochem. Ph.D
Doctoral fellow at catedra de Fisiología-Instituto de Química y Metabolismo del Fármaco (IQUIMEFA-CONICET-UBA)
Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires

Néstor R. Lago, MD
Director of the Center of Experimental and Applied Pathology, Department of Pathology, Faculty of Medicine, University of Buenos Aires, Argentina.
Full Professor of Pathology. Department of Pathology, Faculty of Medicine, University of Buenos Aires, Argentina

Mario Alejandro Lorenzetti, Ph.D
Instituto Multidisciplinario de Investigaciones en Patologías Pediátricas (IMIPP- CONICET-GCBA)
Laboratorio de Biología Molecular, División Patología,
Hospital de Niños Ricardo Gutiérrez, Buenos Aires, Argentina

Ariel R. Marcotegui, MD, MSc
Researcher at the Laboratory of Hepatic Encephalopathy and Portal Hypertension, Department of Pathophysiology, University of Buenos Aires, Ciudad de Buenos Aires, Argentina.

Gustavo A. Moviglia, MD, Ph.D
Director of the Center of Investigation and Engineering of Tissues and Cellular Therapy. Maimonides University. Buenos Aires. Argentina

Alberto E. Muñoz, MD
Hepatologist at the Hospital de Gastroenterología Dr. Carlos B. Udaondo, Buenos Aires Argentina.

Lisandro Orbea, MD. MSc.
Researcher at the Laboratory of Hepatic Encephalopathy and Portal Hypertension.
Faculty of Pharmacy and Biochemistry,
University of Buenos Aires. Buenos Aires, Argentina.
Member of the Center of Experimental and Applied Pathology (CPEA).
Faculty of Medicine, University of Buenos Aires, Buenos Aires, Argentina.

Juan C Perazzo, MD
Director of the Laboratory of Hepatic Encephalopathy and Portal Hypertension
Faculty of Pharmacy and Biochemistry, University of Buenos Aires, Argentina.
Professor of Pathophysiology, Faculty of Pharmacy and Biochemistry, University of Buenos Aires, Argentina.

Luís Perea, MD.
Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS),
Centro de Investigación Biomédica en Red de Enfermedades Hepáticas y Digestivas
(CIBERehd), Barcelona, Spain.

Juan José Poderoso, MD, Ph.D
Director of the Laboratory of Oxygen Metabolism, University Hospital,
School of Medicine, University of Buenos Aires. Argentina
Member of the National Council of Scientific and Technological Research
(CONICET), Argentina
Full Professor of Internal Medicine, Faculty of Medicine,
University of Buenos Aires, Buenos Aires, Argentina

María Victoria Preciado, Ph.D
Instituto Multidisciplinario de Investigaciones en Patologías Pediátricas
(IMIPP- CONICET-GCBA)
Laboratorio de Biología Molecular, División Patología,
Hospital de Niños Ricardo Gutiérrez, Buenos Aires, Argentina.

Myrian R. Rodríguez, Biol. Ph.D
Assistant Researcher at Instituto de Inmunología,
Genética y Metabolismo (INIGEM-CONICET-UBA),
Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires

Salvador Romay, MD
Senior Researcher at the Laboratory of Hepatic Encephalopathy and Portal Hypertension,
Department of Pathophysiology, University of Buenos Aires,
Ciudad de Buenos Aires, Argentina.
Chair of Internal Medicine II, Hospital Pirovano, Ciudad de Buenos Aires, Argentina.

Pau Sancho-Bru, MD. Ph.D
Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS),
Centro de Investigación Biomédica en Red de Enfermedades Hepáticas y Digestivas
(CIBERehd), Barcelona, Spain.

Mercedes Scholler, Msc
Doctoral fellow at catedra de Fisiología-Instituto de Química y Metabolismo del Fármaco
(IQUIMEFA-CONICET-UBA),
Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires

Juan Skerl, MD, MSc
Researcher at the Laboratory of Hepatic Encephalopathy and Portal Hypertension,
Department of Pathophysiology, University of Buenos Aires,
Ciudad de Buenos Aires, Argentina.

Pablo A Souto, Biochem. MSc
Researcher at the Laboratory of Hepatic Encephalopathy and Portal Hypertension.
Faculty of Pharmacy and Biochemistry. University of Buenos Aires.
Buenos Aires, Argentina.

Leandro Steinberg, MD
Gastroentherology Unit Durand Hospital, Ciudad de Buenos Aires, Argentina

Silvina Tallis, Biochem. MSc, Ph.D
Researcher at the Laboratory of Hepatic Encephalopathy and Portal Hypertension,
Department of Pathophysiology, University of Buenos Aires,
Ciudad de Buenos Aires, Argentina.

Júlia Vallverdú
Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS),
Centro de Investigación Biomédica en Red de Enfermedades Hepáticas y Digestivas
(CIBERehd), Barcelona, Spain.

Pamela Valva, MD, Ph.D
Instituto Multidisciplinario de Investigaciones en Patologías Pediátricas
(IMIPP- CONICET-GCBA)
Laboratorio de Biología Molecular, División Patología,
Hospital de Niños Ricardo Gutiérrez, Buenos Aires, Argentina.

Marcelo S. Vatta, Biochem. Ph.D
Professor of Physiology, Physiology Department, Faculty of Pharmacy and Biochemistry,
University of Buenos Aires. Buenos Aires, Argentina.
Principal researcher at Catedra de Fisiología-Instituto De Química Y
Metabolismo Del Fármaco (IQUIMEFA-CONICET-UBA),
Faculty of Pharmacy and Biochemistry, University of Buenos Aires,
Buenos Aires, Argentina.

Soraya Wilke Saliba, Pharm.
Department of Psychiatry, Neurochemistry Lab,
University of Freiburg Medical School, Freiburg, Germany.

Nizar Yousif, MSc
Department of Psychiatry, Neurochemistry Lab,
University of Freiburg Medical School, Freiburg, Germany.

PREFACE

Liver Cirrhosis, similar to congestive heart failure, chronic obstructive pulmonary disease, or chronic kidney disease, represents the final stage of chronic disease which results in the severe and permanent damage of a vital organ. Furthermore, cirrhosis has the peculiarity of being an important risk factor in the development of liver cancer. In recent years liver cirrhosis has emerged as being an enormous health problem worldwide. Global epidemiology studies have shown that the cirrhosis is one of the principal causes of mortality in both men and women. In 2015, the number of deaths attributed to cirrhosis worldwide was almost 1.3 million people. If we then add the 800.000 people who died of liver cancer, which almost always occurs with cirrhosis, the total number of deaths due to a consequence of end stage liver disease surpasses 2 million people. What is more, liver cirrhosis is one of the most important causes of disability-adjusted life years, has a markedly negative impact on patients' quality of life and is also responsible for a high consumption of health resources. For this reason if we are to reduce the impact and mortality of liver diseases in the world, there is a need to increase knowledge and understanding of the pathogenic mechanisms of liver injury and disease progression of chronic liver diseases. This will bring about without doubt an improvement in the early diagnostic methods and treatment of diseases.

In this context, this book by Juan Carlos Perazzo and his co-authors has a special meaning, as it represents a substantial effort in the dissemination of understanding of the biological mechanisms that drive the development and progression of chronic liver diseases. The book covers the different areas from cell biology in the different types of liver cells to the description of the most frequent syndromes in Hepatology. The authors of the chapters have done an excellent job of in-depth reviewing of the various issues without losing sight of the whole picture. For this reason the book is useful for investigators in both experimental and clinical research, as well as clinicians who care for patients with liver disease and want to improve their knowledge and understanding. In this day and age, much of the information is available on the internet but many times it is difficult to select and apply this information. The compilation of this information in a book format is much more attractive as is this book when you hold it in your hands.

Pere Ginès, MD, PhD
Professor and Chairman
Liver Unit, Hospital Clinic of Barcelona
University of Barcelona.

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Broadening our state of consciousness improves human welfare.

This book is an attempt to complete a vision of the chronic liver disease. We focus on the basic molecular mechanisms as a rational basis for the steps that follow, the accurate diagnosis and therapy. In this way, we pretend to achieve a global vision of the organ disease. Many pathophysiological aspects are not included, not because we think they lack of importance, but because we believe that the chosen ones include comprehensive pathophysiological aspects. We emphasize in the interrelationship of the numerous medical disciplines. The impact on an organic system is inextricably linked to its effects on others, but it could be considered that the basic pathophysiological pathways are shared by different pathologies, and each one of them with its own characteristic response. This book is an attempt to expose these shared pathways.

A substantial aspect is that currently the most productible working way is as a team. This book is the result of a team work plus the personal experience that attests it. A broad view also teaches that there are so many people involved in each chapter that is difficult to give a true recognition list. Anyway, we all agree that families and friendship are essential to treat this type of work and that both concepts should be expanded. We recognize and thank all who explicitly and implicitly worked in this presentation.

With the hope that this book could be of some help to researchers and clinicians, we will keep working with the new arriving challenges.

We dedicate this book to our families, whose love, tolerance and support sustained us; to our colleagues, from whom we have learned; to the chapter authors, who have given so much of themselves and, finally, to the readers.

ABSTRACT

Chronic liver disease (CLD) is one of the most prevalent pathologies in developed countries. The large amount of new knowledge drove to subspecialities, even in issues such as hepatology. Every day is more difficult to access all the relevant information that is being published. And even more, biomolecular techniques lead to a complexity level that except of the specific professional working area, becomes so complicated that excludes other professionals.

This book attempts to give a broad overview of the molecular biology of the liver, emphasizing in how this knowledge supports the rationale for treatments. Thus, pathophysiology and therapies are updated in viral hepatitis, hepatic encephalopathy and Portal hypertension, among others. However appended issues, which might look less relevant, as stem cells and endocannabinoids are included. These two issues will be soon relevant due to their close relationship with the liver tissue and especially in liver disease.

New paradigms such as cell death and involvement of the extracellular matrix, are also contemplated. In addition, important issues, such as stellate cells and their intimate relationship with liver function and fibrogenesis are covered in depth.

The basic role of endothelins in CLD is presented in-depth manner. Because of its prevalence, NASH is discussed with special interest, with emphasis in fatty liver process, its molecular relation with liver cells and its metabolism in CLD. We believe this is a broad vision, although it does not cover all issues, which describes the basic pathophysiological mechanisms shared by many liver diseases, giving a rational support for specific therapies.

CONTENTS

In: Chronic Liver Disease: From Molecular Biology to Therapy ISBN: 978-1-53610-237-6
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Chapter 1

CELL DEATH ISSUE FOR RESEARCHERS AND CLINICIANS

Pamela Valva, Mario Alejandro Lorenzetti and María Victoria Preciado

ABSTRACT

Hepatocyte cell death is a central mechanism involved in liver injury and is present in almost all types of human liver diseases. Indeed, excessive cell death has been identified as a central mechanism of liver damage in conditions such as acute and chronic viral hepatitis, alcoholic and non-alcoholic steatohepatitis (ASH and NASH), and drug-induced liver injury (DILI). Different mechanisms of cell death such as apoptosis, necrosis, necroptosis and autophagy, which may vary substantially amongst liver diseases, can trigger specific cell death responses and promote disease progression. In this chapter, we first describe the molecular mechanisms of different forms of liver cell death and then we discuss how cell death contributes to the development of liver disease.

INTRODUCTION

Cell death is likely to be one of the most widely studied topics among cell biologist. It is a well-known fact that tissue homeostasis is dependent on the perfect balance between cell proliferation and cell death. This balance is finely regulated by positive and negative signals, which determine the final fate between life and death; any imbalance in this process may result in disease linked with unwanted cell death or cell growth. Since the initial description of cell death in the 1960s, a number of different death mechanisms have been described (Favaloro et al., 2012). Cell death is defined by the *Nomenclature Committee on Cell Death* as a process by which the cell ceases to carry out its physiological functions, a process that remains reversible until an irreversible phase or 'point-of-no-return' is trespassed. It can be classified according to: 1) the cells morphological appearance (which may be apoptotic, necrotic, autophagic or associated with mitosis), 2) an enzymological criteria (with and without the involvement of nucleases or of distinct classes of proteases, such as caspases,

calpains, cathepsins and transglutaminases), 3) according to functional aspects (programmed or accidental, physiological or pathological) or 4) in relation to immunological outcome (immunogenic or non-immunogenic) (Kroemer et al., 2009). Hepatocyte cell death is a central mechanism involved in liver injury and is present in almost all types of human liver disease. Indeed, excessive cell death has been identified as a central mechanism of liver damage in conditions such as acute and chronic viral hepatitis, alcoholic and non-alcoholic steatohepatitis (ASH and NASH), and drug-induced liver injury (DILI) (Eguchi et al., 2014; Dolganiuc et al., 2012; Bantel and Schulze-Osthoff, 2012; Jaeschke et al., 2012). Clinical data and animal models also suggest that hepatocyte death is the key trigger in liver disease progression, manifested by the subsequent development of inflammation, fibrosis, cirrhosis, and hepatocellular carcinoma (HCC). Different mechanisms of cell death such as apoptosis, necrosis, necroptosis and autophagy, which may vary substantially amongst liver diseases, can trigger specific cell death responses and promote its progression (Luedde et al., 2014). Classically, hepatocyte death has been presented in two mutually exclusive forms: programmed cell death (PCD), or apoptotic cell death, vs. accidental or necrotic cell death, based on morphological criteria. However, a growing understanding of the signalling events involved in triggering cell death has allowed for the dissection of the different molecular pathways that result in hepatocyte death and has even identified a number of cell death modes that present with crosstalk and cooperation in the execution of cell death (Eguchi et al., 2014).

In the healthy liver, cell death controls organ homeostasis, with a tight equilibrium between the loss and replacement of hepatocytes (Michalopoulos and DeFrances, 2005). Turnover is low, with approximately 0.05% of hepatocytes at any given time being removed by apoptosis, mostly in zone 3 (Benedetti et al., 1988). The presence of hepatocyte death, reflected by increased levels of serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST), is the most widely used parameter to screen for and monitor patients with liver disease. Moreover, these markers drive therapeutic decisions, have prognostic value for patients with hepatitis B virus (HBV) and hepatitis C virus (HCV) infections, NASH, and autoimmune hepatitis and correlate with liver-specific mortality in the general population (Feld et al., 2005; Amarapurka et al., 2006; Fassio et al., 2004; Angulo et al., 1999; Hui et al., 2003; Ghany et al., 2009; Lee et al., 2008; Kim et al., 2004; Ruhl and Everhart, 2009). These well-established facts emphasize the importance of cell death as the ultimate driver of liver disease progression and the development of liver fibrosis, cirrhosis, and HCC (Eguchi et al., 2014; Luedde et al., 2014). Notably, the contribution of cell death to liver disease is cell-, stage- and context-specific for instance; whereas increased cell death in hepatocytes contributes to fibrogenesis, cell death in fibrogenic cells is an important mechanism for resolution of liver fibrosis (Luedde et al., 2014; Iredale et al., 1998; Friedman, 2008).

In view of the fundamental role of cell death in virtually all hepatic diseases, precise knowledge on the mechanisms regulating cell death and cell death responses is essential to understand the pathophysiology of liver diseases and to develop new therapeutic approaches.

Forms of Liver Cell Death

Most forms of liver cell death have been extensively characterized *in vitro* in primary hepatocytes cultures or in several immortalized hepatocyte cell lines, but only a few have

been well defined *in vivo* using various experimental animal models or patients with liver diseases. The complexity of studying cellular demise in either *ex vivo* (explanted liver tissue from animal models or liver biopsy tissue from humans) or *in vivo* (model organism and/or humans) comes from the recognition that, in many instances, hepatic cell death represents a highly heterogeneous process. Moreover, frequent overlap and crosstalk between involved pathways may result in molecular transitions between different cell death modalities. In the damaged liver, forms of cell death include apoptosis, necrosis, necroptosis, and autophagy (Figure 1). These medical terms have clear definitions in pathology; however, different modes of cell death are intermingled as a continuous process during liver injury. Therefore, cells initiating a specific form of PCD during an acute or chronic insult could finally evolve into a different form of demise, resulting in a mixed pattern of cell death (Eguchi et al., 2014).

Apoptosis

The term "apoptosis" derives from the Greek meaning "dropping off" and refers to the falling of leaves from the trees in autumn. It is used, in contrast to necrosis, to describe the situation in which a cell actively pursues a course towards its death upon receiving certain stimuli (Kerr et al., 1972). Ever since it was described by Kerr et al. in the 1970's, apoptosis remains one of the most investigated processes in biologic research (Kerr et al., 1972). Apoptosis is a typical form of PCD, an ordered and orchestrated cellular process that occurs in both physiological and pathological conditions. The mechanism of apoptosis is complex and involves many pathways, where any fault, at any given point, may play a pivotal role in the pathogenesis of many diseases.

Under physiological conditions, cell death by apoptosis regulates the sculpting of tissues during embryonic development such as the removal of interdigital webs, shaping of the inner ear or in cardiac morphogenesis. Moreover, in the adult organism, apoptosis regulates involution processes such as shedding of the endometrium, regression of the post-lactating mammary gland, and normal destruction of cells before their replacement (Duprez et al., 2009). Pathological conditions involve for example some forms of virus-induced cell death, such HCV and HBV; pathologic atrophy of organs and tissues such as prostatic atrophy after radiation or hypoxia; degenerative diseases such as Alzheimer's and Parkinson's disease, in immune graft rejection; depletion of $CD4^+$ cells in AIDs; and cell death that occurs in heart diseases such as myocardial infarction (Wong, 2011). Moreover, cancer is one of the scenarios where too little apoptosis occurs, resulting in malignant cells that will not die.

In the liver, apoptosis is particularly important, as this is an organ that is naturally exposed to toxins, drugs, and virus; however, excessive apoptosis can result in tissue destruction and organ failure. Moreover, hepatic apoptosis is considered a prominent pathological feature in most forms of liver injury and usually, an excess in apoptosis of hepatocytes accompanies chronic disease. However, apoptosis may act as a double-edged sword, being the cause of the problem as well as its solution. This fact has impelled many investigations which have now ventured into the quest for new drugs targeting various aspects of apoptosis (Wong, 2011). Interventions in hepatic apoptosis can delay disease progression and reduce the morbidity of liver diseases. Nevertheless, to date, no therapeutic approach has proven successful in clinical practice and those mechanisms responsible for hepatic apoptosis in different liver diseases are still under investigation (Wang, 2015).