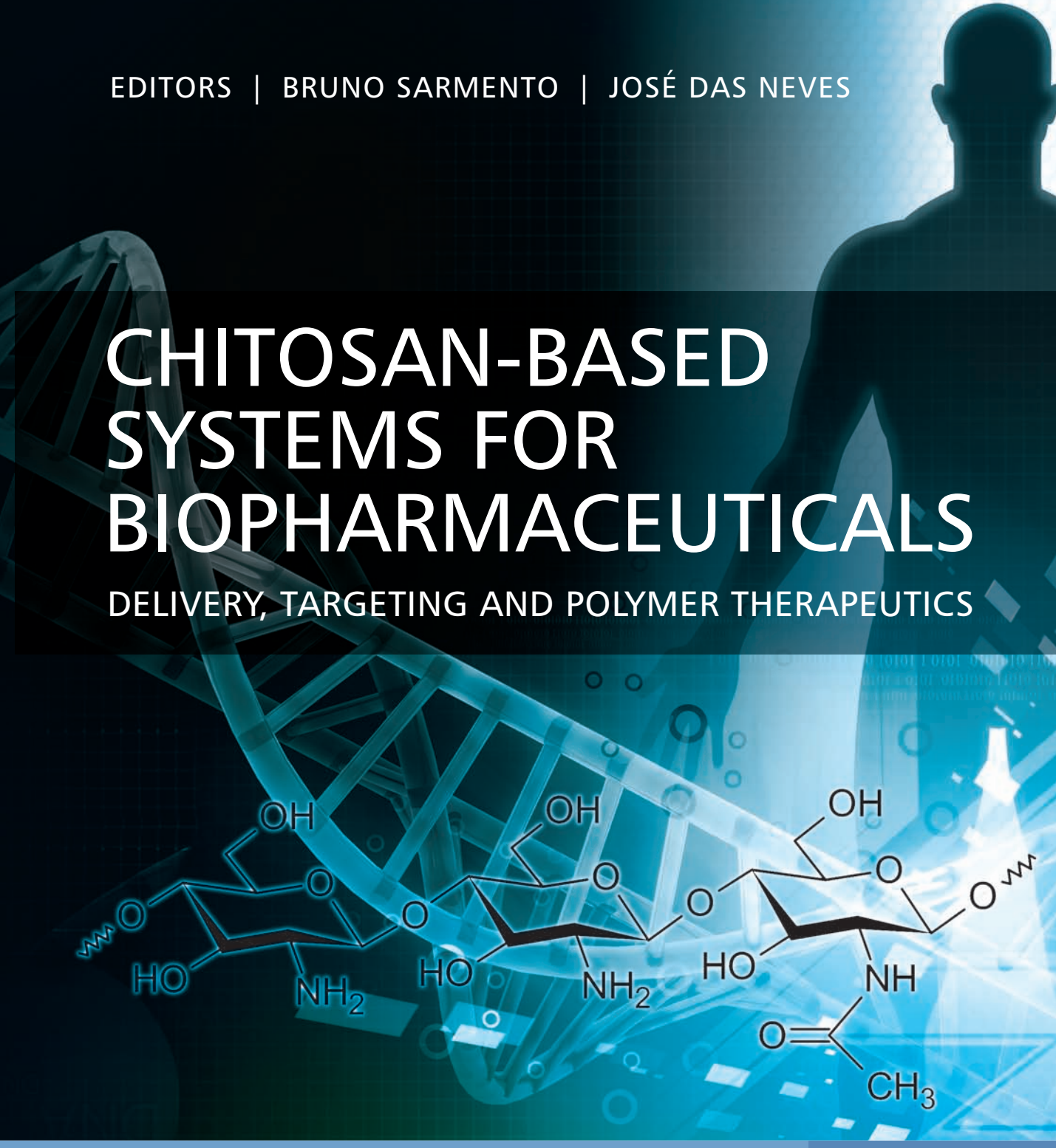
EDITORS | BRUNO SARMENTO | JOSÉ DAS NEVES

CHITOSAN-BASED SYSTEMS FOR BIOPHARMACEUTICALS

DELIVERY, TARGETING AND POLYMER THERAPEUTICS



WILEY

Chitosan-Based Systems for Biopharmaceuticals

Chitosan-Based Systems for Biopharmaceuticals

Delivery, Targeting and Polymer Therapeutics

Edited by

BRUNO SARMENTO

Department of Pharmaceutical Technology, Faculty of Pharmacy, University of Porto, Porto, Portugal
and
CICS, Department of Pharmaceutical Sciences, Instituto Superior de Ciências da Saúde–Norte, Gandra, Portugal

JOSÉ DAS NEVES

Department of Pharmaceutical Technology, Faculty of Pharmacy, University of Porto, Porto, Portugal

WILEY

A John Wiley & Sons, Ltd., Publication

This edition first published 2012

Registered office
John Wiley & Sons Ltd, The Atrium, Southern Gate, Chichester, West Sussex, PO19 8SQ, United Kingdom

Library of Congress Cataloging-in-Publication Data

Chitosan-based systems for biopharmaceuticals : delivery, targeting, and polymer therapeutics / edited by Bruno Sarmento, José das Neves.
p. ; cm.
Includes bibliographical references and index.
ISBN 978-0-470-97832-0 (cloth) – ISBN 978-1-119-96296-0 (ePDF) – ISBN 978-1-119-96297-7 (oBook) – ISBN 978-1-119-96407-0 (ePub) – ISBN 978-1-119-96408-7 (eMobi)
I. Sarmento, Bruno. II. das Neves, José, 1978-
[DNLM: 1. Biopharmaceutics. 2. Chitosan–therapeutic use. 3. Biopolymers–therapeutic use. 4. Drug Carriers–therapeutic use. QU 83]
615.1'9–dc23 2011037452

A catalogue record for this book is available from the British Library.

Print ISBN: 9780470978320

Set in 10/12pt Times by Thomson Digital, Noida, India

Contents

List of Contributors

Wafa I. Abdel-Fattah, Biomaterials Department, National Research Centre, Cairo, Egypt

Toshihiro Akaike, Department of Biomolecular Engineering, Tokyo Institute of Technology, Yokohama, Japan

Marlene Almeida, Hospital de Santo António, Centro Hospitalar do Porto, Porto, Portugal

Sonia Al-Qadi, Department of Pharmacy and Pharmaceutical Technology, University of Santiago de Compostela, Faculty of Pharmacy, Santiago de Compostela, Spain

Mansoor Amiji, Department of Pharmaceutical Sciences, School of Pharmacy, Northeastern University, Boston, MA, USA

Fernanda Andrade, Department of Pharmaceutical Technology, Faculty of Pharmacy, University of Porto, Porto, Portugal

A. Anitha, Amrita Centre for Nanosciences and Molecular Medicine, Amrita Institute of Medical Sciences and Research Centre, Kochi, India

Filipa Antunes, Department of Pharmaceutical Technology, Faculty of Pharmacy, University of Porto, Porto, Portugal

Sambasiva R. Arepalli, Center for Devices and Radiological Health, United States Food and Drug Administration, Silver Spring, MD, USA

Pedro Barrocas, Laboratory of Pharmaceutical Development, R&D Department, Bial - Portela & C.[a], S.A., S. Mamede do Coronado, Portugal

Andreas Bernkop-Schnürch, Department of Pharmaceutical Technology, Institute of Pharmacy, Leopold-Franzens-University Innsbruck, Innsbruck, Austria

M. Cristina Bonferoni, Department of Drug Sciences, School of Pharmacy, University of Pavia, Pavia, Italy

Gerrit Borchard, School of Pharmaceutical Sciences, University of Geneva, University of Lausanne, Geneva, Switzerland

Joel D. Bumgardner, Department of Biomedical Engineering, University of Memphis, TN, USA

Carla M. Caramella, Department of Drug Sciences, School of Pharmacy, University of Pavia, Pavia, Italy

Rui Cerdeira, Laboratory of Pharmaceutical Development, R&D Department, Bial - Portela & C.[a], S.A., S. Mamede do Coronado, Portugal

Chong-Su Cho, Department of Agricultural Biotechnology, and Research Institute for Agriculture and Life Sciences, Seoul National University, Seoul, South Korea

Hee-Jeong Cho, College of Pharmacy, Seoul National University, Seoul, South Korea

Myung-Haing Cho, Laboratory of Toxicology, College of Veterinary Medicine, Seoul National University, Seoul, South Korea

Yun-Jaie Choi, Department of Agricultural Biotechnology, and Research Institute for Agriculture and Life Sciences, Seoul National University, Seoul, South Korea

Teresa Cunha, Hospital de Santo António, Centro Hospitalar do Porto, Porto, Portugal

Tianhong Dai, Wellman Center for Photomedicine, Massachusetts General Hospital, Boston, MA, USA

Department of Dermatology, Harvard Medical School, Boston, MA, USA

José das Neves, Department of Pharmaceutical Technology, Faculty of Pharmacy, University of Porto, Porto, Portugal

Meng Deng, Department of Orthopaedic Surgery, University of Connecticut, Farmington, CT, USA

Gustavo Dias, Hospital de Santo António, Centro Hospitalar do Porto, Porto, Portugal

Michael Dornish, FMC BioPolymer AS/NovaMatrix, Sandvika, Norway

Farnaz Esmaeili, King's College London, Pharmaceutical Science Division, London, United Kingdom

Jonathan Fallon, Laboratory of Tumor Immunology and Biology, National Cancer Institute, CCR, National Institutes of Health, Bethesda, MD, USA

Eduardo Fernandez-Megia, Department of Organic Chemistry and Center for Research in Biological Chemistry and Molecular Materials (CIQUS), University of Santiago de Compostela, Santiago de Compostela, Spain

Franca Ferrari, Department of Drug Sciences, School of Pharmacy, University of Pavia, Pavia, Italy

Masanori Fujita, Research Institute, National Defense Medical College, Saitama, Japan

Qingyu Gao, Institute of Fine Chemical and Engineering, Henan University, Kaifeng, People's Republic of China

Rogério Gaspar, Nanomedicine and Drug Delivery Systems Group, iMed.UL– Research Institute for Medicines and Pharmaceutical Sciences, Faculty of Pharmacy, University of Lisbon, Lisbon, Portugal

John W. Greiner, Laboratory of Tumor Immunology and Biology, National Cancer Institute, Center for Cancer Research, National Institutes of Health, Bethesda, MD, USA

Ana Grenha, Centre for Molecular and Structural Biomedicine, Institute for Biotechnology and Bioengineering, University of Algarve, Faro, Portugal

Maika Gulich, Heppe Medical Chitosan GmbH, Halle (Saale), Germany

Ding-Ding Guo, Department of Agricultural Biotechnology, and Research Institute for Agriculture and Life Sciences, Seoul National University, Seoul, South Korea

Ahmad Sukari Halim, Reconstructive Sciences Unit, School of Medical Sciences, Universiti Sains Malaysia, Kubang Kerian, Kelantan, Malaysia

Michael R. Hamblin, Wellman Center for Photomedicine, Massachusetts General Hospital, Boston, MA, USA

Department of Dermatology, Harvard Medical School, Boston, MA, USA

Harvard–MIT Division of Health Sciences and Technology, Cambridge, MA, USA

Hidemi Hattori, Research Institute, National Defense Medical College, Saitama, Japan

Michael Heffernan, Laboratory of Tumor Immunology and Biology, National Cancer Institute, Center for Cancer Research, National Institutes of Health, Bethesda, MD, USA

Simon Heuking, Vaccine Formulation Laboratory, Department of Biochemistry, University of Lausanne, Epalinges, Switzerland

Ying-Ying Huang, Wellman Center for Photomedicine, Massachusetts General Hospital, Boston, MA, USA

Department of Dermatology, Harvard Medical School, Boston, MA, USA

Aesthetic and Plastic Center of Guangxi Medical University, Nanning, China

Masayuki Ishihara, Research Institute, National Defense Medical College, Saitama, Japan

Shardool Jain, Department of Pharmaceutical Sciences, School of Pharmacy, Northeastern University, Boston, MA, USA

Rangasamy Jayakumar, Amrita Centre for Nanosciences and Molecular Medicine, Amrita Institute of Medical Sciences and Research Centre, Kochi, India

Hu-Lin Jiang, Laboratory of Toxicology, College of Veterinary Medicine, Seoul National University, Seoul, South Korea

Tao Jiang, Zimmer Orthobiologics, Inc., Austin, TX, USA

Yasuhiro Kanatani, Department of Policy Science, National Institute of Public Health, Saitama, Japan

David S. Kaplan, Center for Devices and Radiological Health, US Food and Drug Administration, Silver Spring, MD, USA

Thomas J. Kean, Benaroya Research Institute at Virginia Mason, Seattle, WA, USA

Lim Chin Keong, Reconstructive Sciences Unit, School of Medical Sciences, Universiti Sains Malaysia, Kubang Kerian, Kelantan, Malaysia

Goen Kim, College of Pharmacy, Seoul National University, Seoul, South Korea

You-Kyoung Kim, Department of Agricultural Biotechnology, and Research Institute for Agriculture and Life Sciences, Seoul National University, Seoul, South Korea

Satoko Kishimoto, Research Institute, National Defense Medical College, Saitama, Japan

Research Fellow of the Japan Society for Promotion of Science, Tokyo, Japan

Hyeok-Seung Kwon, College of Pharmacy, Seoul National University, Seoul, South Korea

Cato T. Laurencin, Department of Orthopedic Surgery, University of Connecticut, Farmington, CT, USA

Department of Chemical, Materials and Biomolecular Engineering, University of Connecticut, Storrs, CT, USA

Claus-Michael Lehr, Department of Biopharmaceutics and Pharmaceutical Technology, Saarland University, Saarbrücken, Germany

Helmholtz-Institute for Pharmaceutical Research Saarland (HIPS), Helmholtz Center for Infection Research (HZI), Saarland University, Saarbrücken, Germany

Katharina Leithner, Department of Pharmaceutical Technology, Institute of Pharmacy, Leopold-Franzens-University Innsbruck, Innsbruck, Austria

Brigitta Loretz, Helmholtz-Institute for Pharmaceutical Research Saarland (HIPS), Helmholtz Center for Infection Research (HZI), Saarland University, Saarbrücken, Germany

Riccardo A. A. Muzzarelli, Professor Emeritus of Enzymology, University of Ancona, Ancona, Italy

Shanti V. Nair, Amrita Centre for Nanosciences and Molecular Medicine, Amrita Institute of Medical Sciences and Research Centre, Kochi, India

Ramon Novoa-Carballal, Department of Organic Chemistry and Center for Research in Biological Chemistry and Molecular Materials (CIQUS), University of Santiago de Compostela, Santiago de Compostela, Spain

Yu-Kyoung Oh, College of Pharmacy, Seoul National University, Seoul, South Korea

Rishi Paliwal, Drug Delivery Research Laboratory, Department of Pharmaceutical Sciences, Dr. H. S. Gour Vishwavidyalaya, Sagar, M.P., India

Shivani Rai Paliwal, Drug Delivery Research Laboratory, Department of Pharmaceutical Sciences, Dr. H. S. Gour Vishwavidyalaya, Sagar, M.P., India

Claudia Philippi, Department of Biopharmaceutics and Pharmaceutical Technology, Saarland University, Saarbrücken, Germany

Ahmad Hazri Abdul Rashid, SIRIM, Environmental and Bioprocess Technology Centre, Shah Alam, Selangor, Malaysia

N. Sanoj Rejinold, Amrita Centre for Nanosciences and Molecular Medicine, Amrita Institute of Medical Sciences and Research Centre, Kochi, India

Carmen Remuñán-López, Department of Pharmacy and Pharmaceutical Technology, University of Santiago de Compostela, Faculty of Pharmacy, Santiago de Compostela, Spain

Katja Richter, Heppe Medical Chitosan GmbH, Halle (Saale), Germany

Torsten Richter, Heppe Medical Chitosan GmbH, Halle (Saale), Germany

Ricardo Riguera, Department of Organic Chemistry and Center for Research in Biological Chemistry and Molecular Materials (CIQUS), University of Santiago de Compostela, Santiago de Compostela, Spain

Marguerite Rinaudo, Centre de Recherches sur les Macromolécules Végétales (CERMAV), Centre National de la Recherche Scientifique, affiliated with Joseph Fourier University, Grenoble, France

Silvia Rossi, Department of Drug Sciences, School of Pharmacy, University of Pavia, Pavia, Italy

Giuseppina Sandri, Department of Drug Sciences, School of Pharmacy, University of Pavia, Pavia, Italy

Bárbara Santos, Hospital de Santo António, Centro Hospitalar do Porto, Porto, Portugal

Bruno Sarmento, Department of Pharmaceutical Technology, Faculty of Pharmacy, University of Porto, Porto, Portugal

CICS, Department of Pharmaceutical Sciences, Instituto Superior de Ciências da Saúde–Norte, Gandra, Portugal

Ulrich F. Schaefer, Department of Biopharmaceutics and Pharmaceutical Technology, Saarland University, Saarbrücken, Germany

Tyler G. St. Denis, Wellman Center for Photomedicine, Massachusetts General Hospital, Boston, MA, USA

Columbia University, New York, NY, USA

Branca Teixeira, Hospital de Santo António, Centro Hospitalar do Porto, Porto, Portugal

Maya Thanou, Pharmaceutical Sciences Division, King's College London, London, United Kingdom

Teófilo Vasconcelos, Laboratory of Pharmaceutical Development, R&D Department, Bial - Portela & C.[a], S. A., S. Mamede do Coronado, Portugal

Mafalda Videira, Nanomedicine and Drug Delivery Systems Group, iMed.UL– Research Institute for Medicines and Pharmaceutical Sciences, Faculty of Pharmacy, University of Lisbon, Lisbon, Portugal

Suresh P. Vyas, Drug Delivery Research Laboratory, Department of Pharmaceutical Sciences, Dr. H. S. Gour Vishwavidyalaya, Sagar, M.P., India

Akira Yamamoto, Department of Biopharmaceutics, Kyoto Pharmaceutical University, Kyoto, Japan

Jinfang Yuan, Institute of Fine Chemical and Engineering, Henan University, Kaifeng, People's Republic of China

David A. Zaharoff, Laboratory of Tumor Immunology and Biology, National Cancer Institute, Center for Cancer Research, National Institutes of Health, Bethesda, MD, USA

Biomedical Engineering Program, University of Arkansas, Fayetteville, AR, USA

Ismail Zainol, Chemistry Department, Faculty of Science and Mathematics, Universiti Pendidikan Sultan Idris, Tanjung Malim, Perak, Malaysia

Cuiping Zhai, Institute of Fine Chemical and Engineering, Henan University, Kaifeng, People's Republic of China

Foreword

The reading of the book *Chitosan-Based Systems for Biopharmaceuticals: Delivery, Targeting and Polymer Therapeutics* has given me great pleasure because it represents a nice illustration of the area of research to which I have dedicated an important part of my research career. It was in the early 1990s, working at MIT with Bob Langer on the encapsulation of proteins within poly(lactide-*co*-glycolide) (PLGA) microspheres, that I became conscious of the necessity of new biomaterials for the controlled delivery of delicate compounds, that is, biopharmaceuticals; biomaterials which would be friendly with the associated compounds; biomaterials which could be converted into nanoparticles using mild techniques; and biomaterials that could have a low price based on their wide availability in nature. Chitosan comes to my mind as a wonderful biomaterial fulfilling all these desirable properties. Our goal was to convert chitosan powders into nanoparticles using a procedure that would be adequate for the association of biopharmaceuticals. We were then the first authors reporting the ionotropic gelation technique for the association of proteins to chitosan nanoparticles in 1997. Now, it is amazing for me to see how the history of this biomaterial has evolved. We find thousands of articles and hundreds of patents using the keywords "chitosan nanoparticles." It is, indeed, the biomaterial that has attracted the most significant research attention in the area of nanodrug delivery. As a consequence of this accumulated information, we got to know this unique material quite well. For example, we currently recognize how we can engineer this material in order to make it useful for a variety of interesting biomedical applications and, even more importantly, we can appreciate how this biomaterial is making its way to a final purpose: to provide us with new solutions for improving our health and quality of life.

This book will be of great value to those readers who want to know about chitosan from the perspective of its potential for the delivery of biopharmaceuticals. Following an introductory section, the book is divided in three major parts. The first part is about the general properties of chitosan, with emphasis on the physical–chemical properties that are critical for processing it into adequate delivery systems and also on those of relevance for its use as a biomaterial for human use (biocompatibility and biodegradability). In addition, this part presents the inherent biological properties of chitosan, its behavioral mechanism of action upon contact with living cells and tissues, and the way it interacts with drugs and more precisely with delicate biomolecules such as peptides, proteins, antigens, and nucleic acid-based biocompounds. This part ends by presenting the possibility of chemically modifying chitosan in order to further extend the properties and functionalities of chitosan with regard to its use for the delivery of biopharmaceuticals.

In the second part of the book, the reader will find a great display of the possibilities of chitosan being processed into different pharmaceutical forms, starting by conventional dosage forms and continuing to micro- and nanoparticles. This part logically focuses on the special mucoadhesive properties of chitosan and, thus, on its potential for mucosal drug and vaccine delivery.

The third part is particularly illustrative of the degree of chitosan evolution as a biomaterial. It presents various ways to chemically modify and engineer chitosan in order to make it attractive for a variety of interesting applications, including wound dressing, targeted drug delivery, tissue engineering, and regenerative medicine.

The fourth and final section is without a doubt the most critical one for those who want to know where we stand on the prospects of chitosan as a biomaterial for drug delivery. This section complements the first one regarding the toxicological properties of chitosan under the perspective of the regulatory path and presents the

quality control and good manufacturing practice required for chitosan-derived products. Most significantly, this part covers the amazing information available on chitosan patents and the patentability of chitosan-based biopharmaceutical products, this one being one of the most important applications of chitosan.

Overall, the book presents, in a didactic and well-structured form, critical information for readers interested in the delivery of biopharmaceuticals. It would also be of great benefit for researchers attempting to design, produce, and characterize new biomaterials. It would, of course, also be of interest for any student or researcher interested in the growing field of nanodrug delivery.

María José Alonso
Professor of Biopharmacy and Pharmaceutical Technology
University of Santiago de Compostela (USC), Spain

Preface

Bruno Sarmento[1,2] *and José das Neves*[1]
[1] *Department of Pharmaceutical Technology, Faculty of Pharmacy, University of Porto, Porto, Portugal*
[2] *CICS, Department of Pharmaceutical Sciences, Instituto Superior de Ciências da Saúde–Norte, Gandra, Portugal*

Since the market launch in 1982 of the first recombinant "human" insulin (Humulin®, Eli Lilly, Indianapolis, IN, United States), biopharmaceutical medicinal products have seen a steady rise (with particular boosting in recent years) as important tools of modern therapeutics. With an estimated global market of over $US 167 billion by 2015 [1], biopharmaceuticals are currently widely recognized as highly effective molecules in the management of many metabolic, oncologic, and infectious diseases, as well as in the prevention and *in vivo* diagnosis of such diseases. This particular class of pharmaceuticals is quite heterogeneous and not always clearly defined, comprising different active biological molecules of different complexity such as proteins, peptides, and nucleic acids, among others, which are of biological origin and/or manufactured by biotechnological techniques, usually involving living organisms, cells, or their active components [2]. However, unfavorable physical–chemical properties, poor stability, low permeability, and unsuitable biodistribution of biopharmaceuticals pose important challenges for their adequate pharmaceutical formulation and delivery, and thus their use in therapy. In particular, the challenges in developing adequate materials and systems that allow the use of biopharmaceuticals in daily life are huge. Among the wide variety of proposed solutions for advancing the field [3,4], delivery systems based on chitosan and derivatives have deserved recent singular attention.

The history of chitosan dates back to 1859, when French physiologist Charles Rouget (1824–1904) described the deacetylation of chitin by means of its boiling in the presence of concentrated potassium hydroxide [5]. Immediately, he recognized that the newly obtained product was soluble in acidic solutions, contrasting with the water-insoluble nature of native chitin, thus opening new possibilities for its use. However, it wasn't until 35 years later that the modified chitin received the name "chitosan", which has been attributed to the German physiologist and chemist Felix Hoppe-Seyler (1825–1895) [6]. Nearly one century went by until this modified natural polymer started receiving enough attention as a useful material to be used in the design of drug products [7–9]. Over the years, the study of chitosan revealed that it exhibits several favorable biological properties, such as biocompatibility, biodegradability, low toxicity, and mucoadhesiveness, thus making this polymer a promising candidate for the formulation of biopharmaceuticals. More than a simple excipient for the design of conventional pharmaceutical dosage forms, the development of novel biopharmaceutical delivery systems based on chitosan is a rising subject irrespective of the intended route of administration.

In the present book, renowned experts and researchers from academia, industry, and regulatory bodies provide a concise and up-to-date overview of different issues regarding the application of chitosan and its

derivatives for the development and optimization of biopharmaceutical medicinal products. The book is divided in four different parts. Part One discusses general aspects of chitosan and derivatives, with particular emphasis on issues related to the development of biopharmaceutical chitosan-based systems, comprising a useful background for the following chapters. Part Two deals with the use of chitosan and derivatives in the formulation and delivery of biopharmaceuticals, and focuses on the synergistic effects between chitosan and this particular subset of pharmaceuticals. Further, Part Three continues and complements the previous part by discussing in detail specific applications of chitosan and/or some particular derivatives for biopharmaceutical use. Finally, Part Four presents diverse viewpoints on different issues such as the regulatory, manufacturing, and toxicological requirements of chitosan and its derivatives related to the development of biopharmaceutical products, as well as their patent status and their clinical application and potential.

We expect this book to provide scientists and researchers in the fields of drug delivery, material science, medical science, and bioengineering, as well as professionals in the pharmaceutical, biotechnology, and healthcare industries, with an important compendium of fundamental concepts and practical tools for their daily activities. Also, the broad emphasis on different regulatory issues may turn this book into a relevant starting point for discussion among worldwide regulatory bodies, drug policymakers, and biopharmaceutical companies in pursuing suitable biopharmaceutical products based on chitosan and its derivatives, mostly due to their undoubtedly favorable properties.

References

1. International Market Analysis Research and Consulting Group (2010) Global Biopharmaceutical Market Report (2010–2015).
2. Rader, R.A. (2008) (Re)defining biopharmaceutical. *Nat. Biotechnol.*, **26**, 743–751.
3. Orive, G., Gascon, A.R., Hernandez, R.M. *et al.* (2004) Techniques: new approaches to the delivery of biopharmaceuticals. *Trends Pharmacol. Sci.*, **25**, 382–387.
4. Jorgensen, L. and Nielson, H.M. (2009) *Delivery Technologies for Biopharmaceuticals: Peptides, Proteins, Nucleic Acids and Vaccines*, Wiley, Chichester, West Sussex.
5. Rouget, C. (1859) Des substances amylacées dans les tissus des animaux, spécialement des Articulés (chitine). *C. R. Hebd. Séances Acad. Sci.*, **48**, 792–795.
6. Hoppe-Seyler, F. (1894) Ueber chitin und cellulose. *Ber. Dtsch. Chem. Ges.*, **27**, 3329–3331.
7. Machida, Y. and Nagai, T. (1989) Chitin/chitosan as pharmaceutical excipients, in *Topics in Pharmaceutical Sciences* (eds D.D. Breimer, D.J.A. Crommelin, and K.K. Midha), Fédération Internationale Pharmaceutique, The Hague.
8. Knapczyk, J., Krówczynski, L., Krzek, J. *et al.* (1989) Requirements of chitosan for pharmaceutical and biomedical application, in *Chitin and Chitosan: Sources, Chemistry, Biochemistry, Plysical Properties and Applications* (eds G. Braek-Skjåk, T. Anthonsen, and P. Sandford), Elsevier, London.
9. Illum, L. (1998) Chitosan and its use as a pharmaceutical excipient. *Pharm. Res.*, **15**, 1326–1331.

Acknowledgments

The editors would like to express their deepest gratitude to all the authors for accepting the challenge of writing this work. Also, a special word of appreciation is due to Professor María José Alonso for kindly accepting our invitation to write the foreword, and to everyone at Wiley who assisted in the production of this book.