ELSEVIER
ACADEMIC
PRESS

ADVANCES IN CHEMICAL ENGINEERING

VOLUME 29

MOLECULAR AND CELLULAR FOUNDATIONS OF BIOMATERIALS

ADVANCES IN

CHEMICAL ENGINEERING

Volume 29

Molecular and Cellular Foundations of Biomaterials

Edited by

NICHOLAS A. PEPPAS
Department of Chemical and Biomedical Engineering, and Division of Pharmaceutics, University of Texas, Austin, TX, USA

MICHAEL V. SEFTON
Institute of Biomaterials and Biomedical Engineering, Department of Chemical Engineering and Applied Chemistry, University of Toronto, Ontario, Canada

2004

ELSEVIER
ACADEMIC
PRESS

AMSTERDAM - BOSTON - HEIDELBERG - LONDON - NEW YORK - OXFORD
PARIS - SAN DIEGO - SAN FRANCISCO - SINGAPORE - SYDNEY - TOKYO

ELSEVIER B.V.
Sara Burgerhartstraat 25
P.O.Box 211, 1000 AE
Amsterdam, The Netherlands

ELSEVIER Inc.
525 B Street, Suite 1900
San Diego, CA 92101-4495
USA

ELSEVIER Ltd
The Boulevard, Langford Lane
Kidlington, Oxford OX5 1GB
UK

ELSEVIER Ltd
84 Theobalds Road
London WC1X 8RR
UK

First edition 2004

ISBN: 0-12-008529-1
ISSN: 0065-2377 (Series)

♾ The paper used in this publication meets the requirements of ANSI/NISO Z39.48-1992 (Permanence of Paper).
Printed in Great Britain.

CONTRIBUTORS

Numbers in parentheses indicates the pages on which the authors' contribution begin.

KRISTI S. ANSETH, *Howard Hughes Medical Institute, Department of Chemical and Biological Engineering, University of Colorado, Boulder, CO 80309-0424, USA* (7)

PETR BURES, *Departments of Chemical and Biomedical Engineering, and Division of Pharmaceutics, CPE 3.466, The University of Texas at Austin, Austin, TX 78712-0231, USA* (75)

THOMAS D. DZIUBLA, *Institute for Environmental Medicine, University of Pennsylvania School of Medicine, Philadelphia, PA 19104, USA* (75)

F. KURTIS KASPER, *Department of Bioengineering, Rice University, Houston, TX 77005, USA* (131)

MATT J. KIPPER, *Department of Chemical Engineering, Iowa State University, Ames, IA 50011, USA* (169)

ANTHONY M. LOWMAN, *Department of Chemical Engineering, Drexel University, Philadelphia, PA 19104, USA* (75)

SURYA K. MALLAPRAGADA, *Department of Chemical Engineering, Iowa State University, Ames, IA 50011-2230, USA* (47)

KRISTYN S. MASTERS, *Howard Hughes Medical Institute, Department of Chemical and Biological Engineering, University of Colorado, Boulder, CO 80309, USA* (7)

ANTONIOS G. MIKOS, *Department of Bioengineering, Rice University, Houston, TX 77005, USA* (131)

BALAJI NARASIMHAN, *Department of Chemical Engineering, Iowa State University, Ames, IA 50011, USA* (169)

NICHOLAS A. PEPPAS, *Departments of Chemical and Biomedical Engineering, and Division of Pharmaceutics, CPE 3.466, The University of Texas at Austin, Austin, TX 78712-0231, USA* (75)

JENNIFER B. RECKNOR, *Department of Chemical Engineering, Iowa State University, Ames, IA 50011-2230, USA* (47)

MICHAEL V. SEFTON, *Institute of Biomaterials and Biomedical Engineering, Department of Chemical Engineering and Applied Chemistry, University of Toronto, Toronto, Ontario, M5S 3G9 Canada* (1)

PREFACE

Over the past forty years the field of Biomaterials Science and Engineering has grown from a small research area of no more than twenty researchers worldwide, to a robust discipline that has become a cornerstone of the field of Biomedical Engineering. During this time period, the field of biomaterials has found a welcome home in academic chemical engineering departments and in companies working with artificial organs, medical devices, and pharmaceutical formulations. The contributions of chemical engineers to the definition and the growth of the field have been important and at times seminal. It was therefore only natural for us to edit a volume that would highlight some of the major contributions of the chemical engineering world to biomaterials science and engineering.

In the mid 1960s biomaterials science was still at its infancy. The development of biomaterials was an evolving process. As Robert Langer of MIT and I indicated in a recent article (AIChE Journal, 49, 2990 (2003)), many biomaterials in clinical use were not originally designed as such but were off-the-shelf materials that clinicians found useful in solving a problem. Thus, dialysis tubing was originally made of cellulose acetate, a commodity plastic. The polymers initially used in vascular grafts, such as Dacron, were derived from textiles. The materials used for artificial hearts were originally based on commercial-grade polyurethanes. These materials allowed serious medical problems to be addressed. Yet, they also introduced complications. Dialysis tubing would activate platelets and the complement system. Dacron-based vascular grafts could only be used if their diameter exceeded about 6 mm. Otherwise occlusion could occur because of biological reactions at the blood–material and tissue–material interfaces. Blood–materials interactions could also lead to clot formation in an artificial heart, with the subsequent possibility of stroke and other complications.

In the last few years, novel synthetic techniques have been used to impart desirable chemical, physical, and biological properties to biomaterials. Materials have been synthesized either directly, so that desirable chain segments or functional groups are built into the material, or indirectly, by chemical modification of existing structures to add desirable segments or functional groups. It is possible to produce polymers containing specific hydrophilic or hydrophobic entities, biodegradable repeating units, or multifunctional structures that can become points for three-dimensional expansion of networks. Another synthetic approach involves genetic

engineering for the preparation of artificial proteins of uniform structure. This enables the synthesis of periodic polypeptides that form well-defined lamellar crystals, polypeptides containing non-natural amino acids, and monodisperse helical rods. Important issues to be addressed include immunogenicity and purification from contaminants during large-scale production. If techniques were developed to produce polymers with the use of non-amide backbones, the versatility of this approach would be extended.

In this volume, we have collected a series of important critical articles on the present and future of biomaterials science as viewed by some of the leading chemical engineers of the field. It was not our intention to cover all aspects of biomaterials science but rather to unify certain synthetic, structural, and biological topics, and to point out the significant contributions of chemical engineers to the field. It is not a coincidence that this book is part of the well-known series of *Advances in Chemical Engineering*.

As I was commissioning the various chapters included in this volume, I wanted to highlight the main directions of this field: (i) novel methods of synthesis; (ii) advanced design; (iii) advanced characterization methods; (iv) better understanding of biomaterials/tissue interactions; and (v) a wealth of applications. Concerning this last point, it must be noted that just 25 years ago, the term biomaterials referred to materials in contact with the body but was restricted to materials for artificial organs and extracorporeal devices. The "explosion" of the fields of drug delivery and tissue engineering has led to new function and applications of biomaterials. The use of biomaterials in nanoscale technology requires added appreciation for the importance of chemical engineering principles in biomaterials science and engineering.

After a masterful introduction of the field and its new directions by Michael Sefton of the University of Toronto, Kristi Anseth of the University of Colorado offers a critical analysis of cell–materials interaction problems with emphasis on the nature of cell adhesions, adhesion ligands, and surface chemistry.

Surya Mallapragada of Iowa State University addresses questions related to the use of biomaterials in tissue engineering and nerve regeneration, while Anthony Lowman of Drexel University offers a detailed structural analysis of biological hydrogels used in biomaterials and drug delivery applications. Antonios Mikos of Rice University offers a critical review of biomaterials for gene therapy, whereas Balaji Narasimhan of Iowa State University pursues the question of biodegradability in materials, especially those used as drug delivery carriers.

As you read this book, I hope you will appreciate the infinite possibilities of biomaterials science in solving important medical problems.

If this book can influence young engineers and scientists to pursue a career in biomaterials science and engineering, it will have made a lasting impact. I want to thank Michael Sefton for coming to this project with an open mind and adding his advice as a co-editor and author of the first chapter. And I am indebted to the two early chemical engineering giants of the field, Edward Merrill of MIT and Alan Hoffman of the University of Washington, for having taken the first giant leaps in the tortuous road that is "biomaterials".

NICHOLAS A. PEPPAS
AUSTIN, TEXAS, USA
March 2004

CONTENTS

Polymeric Biomaterials for Nerve Regeneration

SURYA K. MALLAPRAGADA AND JENNIFER B. RECKNOR

Structural and Dynamic Response of Neutral and Intelligent Networks in Biomedical Environments

ANTHONY M. LOWMAN, THOMAS D. DZIUBLA, PETR BURES, AND NICHOLAS A. PEPPAS

Biomaterials and Gene Therapy

F. KURTIS KASPER AND ANTONIOS G. MIKOS

Surface-Erodible Biomaterials for Drug Delivery

BALAJI NARASIMHAN AND MATT J. KIPPER

THE NEW NEW BIOMATERIALS

Michael V. Sefton

Institute of Biomaterials and Biomedical Engineering, Department of Chemical Engineering and Applied Chemistry, University of Toronto, 4 Taddle Creek Rd, Suite 407, Toronto, Ontario, M5S 3G9 Canada
E-mail: sefton@chem-eng.utoronto.ca

I. What Happened to Inert Biomaterials?

In the beginning, there were metals and materials scientists. Plastics, polymers, and soft materials came later and then came the chemical engineers. The artificial heart program had a few (Artificial Heart Program Conference, 1969) but it was the artificial kidney program and the interest in new membranes that really started things off. Merrill at Massachusetts Institute of Technology (Merrill *et al.*, 1966) was a pioneer as was Leonard and Gregor (Friedman *et al.*, 1970) at Columbia and Hoffman at the University of Washington. Now almost every chemical engineering department has someone working on biomaterials or there is a bioengineering department nearby with chemical engineers on faculty. Several illustrations of this activity are apparent in this volume.

In the beginning the emphasis was on biocompatibility. Inertness was the key. We had our lists of no's (Table I) and the paradigm was focused on finding, synthesizing or surface modifying materials to make them fit these negative commandments. Interestingly, a large part of the early involvement of chemical engineers was to make materials that were not inert. Heparin immobilization was a hot topic in the late sixties and early seventies and the whole purpose was to make a surface that would actively interact with blood and prevent clotting. "Anti-thrombogenicity" was the keyword.

Advances in Chemical Engineering, vol. 29
ISSN 0065 2377
DOI 10.1016/S0065-2377(03)29001-3

TABLE I

Commandments for inert biomaterials
No toxicity
No hemolysis
No pyrogens (endotoxin)
No protein or cell consumption
No thrombosis (and no emboli)
No inflammation
No infection
No immune response
No complement activation
No carcinogenicity and mutagenicity

Now with tissue engineering, regenerative medicine and combination products, active materials are the topic of interest of biomaterials specialists.

Some active materials are carriers for drugs (drug delivery systems), some have immobilized peptides to enable cell adhesion or migration, some are degradable by hydrolysis or by specific enzyme action. Some contain bioactive agents (e.g., heparin, thrombomodulin) to prevent coagulation or platelet activation while others incorporate bioactive groups to enhance osteoconduction. Many include polyethylene oxide to retard protein adsorption and this is perhaps the closest we have come to a kind of inertness.

The advent of these materials has challenged the regulatory authorities since the materials are no longer being used simply for medical devices. Some include drugs and some include cells or biologicals. It was once sufficient to show that the material had no effect (i.e., it was inert) and then to get the blessing of the regulatory authorities. Now, it is the presence of an effect and a significant one at that, that needs to be regulated. The FDA has established an Office of Combination Products (http://www.fda.gov/oc/combination/) to deal with these products and every indication suggests that it is not long before these products are the norm. It is now not so simple to argue that the next generation of medical devices "does not achieve any of its primary intended purposes through chemical action within or on the body of man" as it is given in part of the FDA definition of a medical device.

II. Biocompatibility of Modern Biomaterials

When biomaterials were inert it was simple to think of biomaterials in terms of the absence of inflammation or the absence of thrombi. Now, with

these newer combination materials we think of biocompatibility in more complex and subtle terms. The "appropriate host response" associated with the definition of biocompatibility has much more subtlety and complexity than we had hitherto considered. Blood compatibility may require some limited platelet adhesion and activation to passivate a material rather than the complete absence of adherent platelet deposits, especially if we want to limit embolization. What we now really mean by blood compatibility has been described in more detail elsewhere (Sefton *et al.*, 2000).

We now recognize that blood compatibility is more complex than it was because we have to consider more than just platelets and coagulation factors and we have to consider the interactions among all the components of blood, including neutrophils, monocytes, and complement. This has led to the conclusion that thrombogenicity is really a special case of inflammation. That modern hematologists disregard Factor XII and the intrinsic coagulation system and focus on tissue factor (Jesty *et al.*, 1995) and that tissue factor is expressed on activated monocytes (Gorbet *et al.*, 2001) highlights further this linking of thrombogenicity and inflammation.

More fundamentally though the performance of these new biomaterials is challenging the entire concept of biocompatibility. A scaffold that promotes cell invasion may contain many of the attributes that in another context would lead to inflammation. Some constructs rely on a limited degree of inflammation to generate the enzymes that will cause the desired remodeling of the construct. Other uses of a biomaterial (e.g., as a vaccine adjuvant) is based on generating a local inflammatory response in order to boost the immune response, while immune responses to tissue constructs is an important, yet largely overlooked, element of the host response (Babensee *et al.*, 1998). Some new angiogenic biomaterials (Gorbet *et al.*, 2003) are designed to control the functional diversity of the monocyte (Riches, 1995), enabling a pro-angiogenic phenotype to emerge as the dominant functional form of these cells. The result is monocyte activation, but "good" activation: producing the blood vessels associated with granulation tissue but without the undesirable cytokines and other inflammatory mediators and proliferating fibroblasts. These new biomaterials are leading us to ask whether inflammation is bad or whether a little bit of inflammation can be a good thing?

Biomaterials are solid drugs. Rather than thinking of biomaterials as an inert contributor, my laboratory has taken to thinking about biomaterials as agonists of a biological response, much like drugs. However, biomaterials are solids and interact with cells and tissues through an interface, making the study of biomaterials more difficult than that of drugs, which are one-dimensional compared to the three-dimensional

biomaterial. The biological responses we are interested in range from protein adsorption and platelet activation but extend to angiogenesis, matrix metalloproteinase secretion, immune recognition, and a wide variety of other biological phenomena. We can make use of the wealth of information, reagents, and assays that are available on these phenomena, but it is necessary to adapt them for the complexities of the interfaces and the differences between drugs and biologically active materials (Table II).

The differences in Table II are intended as broad generalities and readers can easily come up with exceptions or questions about what is meant by a biologically active material. For example, is the action of a drug delivery device always "local" or is a nanoparticle "large" and a DNA drug "small." Thus these characteristics must be interpreted and ringed with qualifiers to be strictly correct.

TABLE II

Biologically active materials	Drugs
Large, 3D objects	Small, 1D molecules
Immobile	Diffusible
Action is local	Action may be systemic, with side-effects a critical concern
Subject to foreign body reaction, coagulation, complement activation, etc.	Inflammation rarely a consideration
Interact across a cell membrane although endocytosis may occur	Act through a cell surface receptor or intracellularly
Limited surface area and ligand density	Even at nanomolar levels, there are many, many ligands (excess ligands?)
Action is often nonspecific	Specificity is key element
Protein adsorption influences cell response through altered ligand or receptor presentation or changes to microenvironment	No equivalent concept, although cell microenvironment affects drug action
Metabolism rarely relevant	Metabolized after an effect or to actually generate the effect
Effect is chronic	Effect is generally short-lived (half-life is a critical parameter)
Effect is generally permanent—pharmacokinetics and bioavailability are not normally considered	Effect is generally not permanent—pharmacokinetics and bioavailability are important
Can be engineered to be degradable and eliminated but many are not	Drug elimination is critical element of design

One of the more troubling characteristics is that of "specificity." Certainly a material that contains an immobilized growth factor or enzyme, contains much of the specificity of the immobilized agent. However, here I am thinking more about the biomaterial that has bioactivity (e.g., angiogenesis or osteoconduction), but without the obvious therapeutic agent within it. Here, the effect appears to be more nonspecific than that seen with drugs. This has been controversial, especially when presented in the form that many materials act the same (with occasional and important exceptions) resulting in questioning the importance of surface chemistry differences among materials (Sefton *et al.*, 2001a). The implications of this with respect to hemocompatibility testing has also been discussed in reference Sefton *et al.*, (2001b). The absence of substantive differences in platelet and leukocyte activation among many materials (Sefton *et al.*, 2001a) suggests that the mechanism of these responses is fundamentally nonspecific in character.

The host response central to biocompatibility is to a 3D object, the chemistry of which does not appear to be terribly important. One way of thinking about this is that the biology does not really care if one changes the chemistry of a surface from one kind of nonspecific surface to another. Only when specificity is introduced through some sort of deliberate design can the biology "appreciate" what is happening. Hence it is little surprising that biomaterials specialists in 2003 speak of understanding the mechanism of biological response as much as they may tout a novel biomaterial. There is an extensive biological literature that we have only started to appreciate and exploit. The prospects for further basic research in biomaterials is correspondingly strong.

REFERENCES

Artificial Heart Program Conference; proceedings, Washington, D.C., June 9–13, 1969. Edited by Ruth Johnsson Hegyeli. National Institutes of Health.

Babensee, J. E., Anderson, J. M., McIntire, L. V., and Mikos, A. G., Host response to tissue engineered devices. *Adv. Drug Deliv. Rev.* **33,** 111–139 (1998).

Friedman, L. I., Liem, H., Grabowski, E. F., Leonard, E. F., and McCord, C. W., Inconsequentiality of surface properties for initial platelet adhesion. *Transactions—American Society for Artificial Internal Organs* **16,** 63–73 (1970).

Gorbet, M. B., and Sefton, M. V., Expression of procoagulant activities on leukocytes following contact with polystyrene and PEG grafted polystyrene beads. *J. Lab. Clin. Med.* **137,** 345–355 (2001).

Gorbet, M., Eckhaus*, A., Lawson-Smith, R., May, M., Sefton, M., and Skarja, G., Material-induced angiogenesis in impaired wound healing. Wound healing Society Abstract, May 2003.

Jesty, J., and Nemerson, Y., The pathways of blood coagulation, *in* "Williams Hematology" (E. Beutler, M. A. Lichtman, B. S. Coller, and T. J. Kipps Eds.), 5th Edition, pp. 1227–1238. McGraw-Hill, New York (1995).

Merrill, E. W., Salzman, E. W., Lipps, B. J., Jr., Gilliland, E. R., Austen, W. G., and Joison, J., Antithrombogenic cellulose membranes for blood dialysis. *Transactions—American Society for Artificial Internal Organs* **12,** 139–150 (1966).

Riches, D. W., Signalling heterogeneity as a contributing factor in macrophage functional diversity. *Semin. Cell Biol.* **6**(6), 377–384 (1995).

Sefton, M. V., Gemmell, Cynthia, H., and Gorbet, Maud B., What really is blood compatibility? *J. Biomater. Sci. Polymer Ed.* **11**, 1165–1182 (2000).

Sefton, M. V., Sawyer, A., Gorbet, M., Black, J. P., Cheng, E., Gemmell, C., and Pottinger-Cooper, E., "Does surface chemistry affect thrombogenicity of surface modified polymers?". *J. Biomed. Mater. Res.* **55,** 447–459 (2001a).

Sefton, M. V., Perspective on hemocompatibility testing. *J. Biomed. Mater. Res.* **55,** 445–446 (2001b).

CELL–MATERIAL INTERACTIONS

Kristyn S. Masters* and Kristi S. Anseth

Howard Hughes Medical Institute, Department of Chemical and Biological Engineering, University of Colorado, Boulder CO 80309, USA

I. Introduction

The nature of cell adhesion to substrate materials has a tremendous effect on cell function and tissue development. Signaling cascades initiated by cell adhesion have the ability to regulate a variety of events, including embryogenesis, tissue differentiation, and cell migration (Koenig and Grainger, 2002; Longhurst and Jennings, 1998). Signaling via receptor–ligand interactions provides the cell with vital information about its

*E-mail: kristi.anseth@colorado.edu

Advances in Chemical Engineering, vol. 29
ISSN 0065 2377
DOI 10.1016/S0065-2377(03)29002-5