

livestock are discussed, because the introduction of rearranged immunoglobulin genes into livestock is a major area of research. All in all, however, the explanation of knockout and replacement technology is quite valuable for the newcomer.

The production of transgenic livestock is an exciting area, and it is handled very well in the next chapter. The current status of this field is well described, and the management of some of these larger species, even if a bit more detailed than necessary, makes entertaining reading. Obviously, the enormous cost of producing these animals is a major problem. The perfection of other methods of gene transfer, and/or of embryonic stem cell technology, would contribute significantly to the further advancement of this field, though microinjection clearly works well.

The final chapter, on 'minor transgenic systems' was perhaps included for the sake of completeness. These systems are quite disparate, and relatively little work has yet been done in many of them.

Nonetheless this information is useful because it satisfies potential curiosity and provides a reference base in the event one or more of these areas develops substantially.

Overall, this book covers a very large area of science surprisingly well. For an individual with extensive knowledge of one transgenic field but little understanding of others, it makes enjoyable reading. It also would serve as nice introductory reading for the graduate student or advanced undergraduate. Where possible the book is comprehensive, but where this is not possible, the book does not lapse into prosaic, detailed discussions of specific experiments that would inevitably lead to an imbalanced presentation. That a readable, informative book could be distilled from the mass of information available in this field is a credit to the authors and editor.

Jan. W. Gordon

**Patch-Clamp Applications and Protocols**, edited by A.A. Boulton, G.B. Baker and W. Walz, 1995. Humana Press Inc. Totowa, NJ, USA, 1995. viii + 316 pp. \$89.50. ISBN 0-89603-311-2.

Written by leading experts from around the world, this book comprehensively reviews the latest on the patch-clamp technique. Contributors offer practical advice on how to overcome the problems encountered in performing and analyzing patch-clamp experiments. Most of the different chapters provide all the information needed to set up and carry out any of the procedures without having to use other resource books.

This book can be divided into three sections. The first section summarizes some basic concepts on the fabrication and properties of patch pipets, cell-free ion channel recordings and on whole-cell patch-clamp recordings. The chapter on 'Patch-Clamp Electrode Technology' reviews, in great detail, the methods used to obtain low-noise whole-cell or single channel recordings. A particular emphasis is given to the choice of the glass used in electrode fabrication. The chapter on 'Cell-free Ion-Channel Recording' gives an excellent description of the methods and problems encountered in performing inside-out membrane patches (like problems of channel rundown and problems of diffusion to the ion channel in the patch); it also provides a list of tips and hints for performing a successful experiment.

The second section contains more specialized applications of the patch-clamp technique. There are three chapters that illustrate the use of different methods of controlling the composition and concentration of the solutions inside and outside the patch-pipet. The chapter on the perforated patch-clamp technique gives a brief but informative review on the different protocols (ATP, nystatin, amphotericin) used to

permeabilize the membrane patch. Hamill and McBride describe a novel variant of the patch-clamp technique: the pressure clamp method. This method allows the application of precise and rapid suction or pressure steps to membrane patches and whole cells and has been recently used to study mechano-gated ion channels. The chapter on the 'Loose Patch-Clamp technique' gives a useful overview on the technique, showing its unique features over other methods and pointing out possible sources of errors and artifacts. There is also a chapter devoted to the use of the patch-clamp technique on more intact preparations such as brain slices.

The last section contains protocols on the combination of patch-clamp and molecular biological techniques. The chapter on single-cell RT-PCR gives a detailed description of the technical steps involved in this procedure. The authors show that the presence of different types of mRNA (and splice variants) can be analyzed within a cell after patch-clamp recording and, in some instances, correlated with the functional properties of the cell. The chapter on receptor/ion-channel expression gives hands-on procedures on oocyte DNA or RNA microinjection, two-electrode voltage clamp and oocyte patch-clamp.

Overall, this book will be an extremely useful tool for electrophysiologists and advanced students who wish to acquire practical skills and first-hand experience in the patch-clamp technique.

Andrés F. Oberhauser

**Introduction to Biocatalysis Using Enzymes and Micro-organisms**; Edited by S.M. Roberts, N.J. Turner, A.J. Willetts and M.K. Turner, Cambridge University Press, Cambridge, 1995, xii + 197 pp. ISBN 0-521-43070-4.

This timely and comprehensive introduction to biocatalysis not only provides the reader with historical perspective and the current state of the art, but addresses issues and challenges pertinent to the practitioner. Biocatalysis has evolved as a very interdisciplinary field, encompassing microbial biochemistry, enzymology, organic chemistry, and chemical engineering. More recently, the tools of recombinant DNA technology have added significantly to the field. This development has greatly expanded the range of enzymes available for study, the ease of studying new enzymes, and the potential to engineer novel features into existing biocatalysts. Overviewing this interdisciplinary and rapidly expanding field for the non-practitioner presents a unique challenge to the authors. Furthermore, since academic and industrial practitioners bring different perspectives and focus to this field, a comprehensive treatise needs to balance contributions from these sectors. These four experienced and well known scientists have handled this challenge very well.

The text consists of six chapters divided into four sections: a historical perspective (Chapter 1); an introduction to biocatalytic systems available and how to choose them (Chapter 2); reviews of biocatalyzed reactions arranged by three general reaction types –

hydrolytic, redox, and C–X bond forming (Chapters 3–5); and industrial scale applications of biocatalysis (Chapter 6).

In their introductory chapter, the authors place the recent surge of interest in biocatalysis (since the mid 80's) in proper historical perspective as a 'reconvergence' of the fields of synthetic organic chemistry and biochemistry which had diverged earlier to pursue separate agendas. Various examples of key biochemical technology developed early in the century are highlighted, including several which integrate biotransformations and organic synthesis. The five-step Reichstein process, developed in the 1930's for the synthesis of ascorbic acid, remains today as the primary industrial process for this important vitamin.

The second chapter addresses many of the practical aspects of biocatalysis, and is geared towards the 'non-microbiologist'. Topics include enzyme classification, enzyme cofactors and cofactor recycling, criteria for choosing between a whole cell versus 'free' enzyme system, and immobilization of enzymes and cells. The authors use the well studied Baeyer–Villiger type biooxidation to outline how a biocatalyst is chosen, where microorganisms can be obtained, and how they are grown, induced, and optimized for a given biotransformation. Since

topics covered in this chapter might easily constitute a text alone, an extensive list of review articles is provided at the end of the chapter for further reading.

The separation of chapters 3–5 based on three reaction types is logical, and is clearly connected to considerations detailed in Chapter 2. The authors have resisted the temptation to review, choosing instead to cite case studies which collectively demonstrate the scope of reactions studied to date. Elaboration is left to the reader with recent reviews cited at the end of each chapter.

The final chapter is chronologically a continuation of Chapter 1, and outlines industrial biocatalytic processes in use today for the production of amino acids, steroids, vitamins, antibiotics, and fine chemicals. The authors also have included recent developments which tend to lie outside the traditional fermentation arena, such as the production of acrylamide, chiral intermediates such as (S)-2-chloropropionic acid, biopolymers (e.g. polyesters), and complex polysaccharides. A promising new direction in the field, the application of enzymes outside normal operating ranges of temperature, pH, and aqueous reaction media is addressed briefly. The chapter finishes with

a description of the Genentech process for ascorbic acid, a unique example of how metabolic and genetic engineering tools are combined to prepare a biocatalyst.

This book would lend itself well both as an introductory textbook and as a teaching resource. Bibliographies at the end of each chapter allow expansion of particular topics. It is particularly satisfying to see a balanced treatment of both whole cell biocatalysis (biotransformation) and enzyme catalyzed synthesis based on commercially available enzymes in a single text. Too often, earlier texts and reviews have treated these research areas separately to target a particular audience. The authors have also been careful to note the potential for cleaner chemical processes, but have not oversold the 'green' aspects of biocatalysis. Some editing errors in chemical structures can be found in Chapters 3–5, but are limited to a missing bond or methylene group and do not hinder understanding of the reactions. Overall, *Introduction to Biocatalysis Using Enzymes and Micro-organisms* fills an important instructional need, providing a balanced overview of a complex field.

Paul Swanson

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**Protein–Solvent Interactions;** Edited by Roger B. Gregory; Marcel Dekker, New York, USA, 1994; xix + 570 pp. \$185.00. ISBN 0-89603-301-5.

The aim of this book could not be stated more clearly than in the editor's preface: 'Our understanding of protein–solvent interactions has ... over the last 15 years ... been marked by a number of interesting and important discoveries. The contributions in this volume present some of these advances in our understanding of structural, thermodynamic and dynamic aspects of protein–solvent interactions. It is not intended to be a comprehensive survey, but the chosen topics will give some sense of the real progress achieved and the outstanding questions that must be addressed.'

The editor's modest disclaimer is belied by the scope of the chapter titles; the book is a broad and much-needed survey of biophysical aspects of the interplay between proteins and their solvent. On the structural side, a chapter deals systematically with bound water molecules that have been located by X-ray crystallography of proteins, summarising the relevant principles that emerge from the 500 known protein crystal structures. Two chapters treat the hydration of proteins at low water:protein ratios; this topic will seem obscure to many biochemists, but in fact it gives important information about water tightly associated with the polypeptide and its side-chains, and it is therefore relevant for proteins in solution, where a tightly-bound water layer is still present on the protein's surface. Rather more specialised chapters include the study of solvent–protein-interaction by scattering of Mössbauer radiation (it is a pity that the type-setter's otherwise apparently inexhaustible stock of symbols did not include the *Umlaut*); a case study in ligand-binding kinetics (the myoglobin–CO reaction investigated by flash photolysis); and a review of the physical principles governing precipitation-based separation of proteins. Of perhaps more general interest are a chapter on the effects of solvent viscosity upon protein dynamics, which places protein dynamics in the context of modern kinetic theory; one on the influence of solvent upon protein stability and association, which attempts to demarcate the 'missing information' still needed for a statistical-mechanical description of these phenomena; a review of thermodynamic mechanisms responsible for the well-known but little-understood enthalpy/entropy compensation effect; and a review of the interaction between proteins and water containing co-solvents. Predictably, all the proteins discussed are globular, and almost all the solvent systems are aqueous: the book purports to be a state-of-the-art review, and such is the state of the art. The chapter on proteins in non-aqueous environments is by far the book's shortest.

Inevitably, a weakness of thermodynamic treatments of proteins in solution is the use of the standard state of infinite dilution, which is highly unphysiological. A chapter on thermodynamic non-ideality and protein solvation presents a well-argued case for the equivalence of classical and statistical-mechanical treatments of these topics; it is emphasized that the statistical-mechanical view is the more productive. This chapter revises some earlier misconceptions in both the classical and the statistical-mechanical treatments and may become a milestone contribution to a general re-assessment of non-ideal solutions of macromolecules.

141 of the book's 547 pages are devoted to a long chapter with the eye-catching title 'The New Paradigm for Protein Research'. This chapter, comprising > 25% of the book's length and therefore costing the purchaser some fifty bucks, argues that the way ahead for protein research is the 'knot-matrix' principle proposed by Gregory and Lumry in 1985. The discourse includes a sweeping review of physical aspects of protein structure, and the author of the chapter (Lumry) has understandably been allotted by the editor of the book (Gregory) the space to expound his views in depth. Whether these views are right, the reader and history will judge; there is certainly a case to be answered. However, the paradigm shift has yet to make a deep impression, as the index entries under 'knot' lead only to allusions by these two contributors. This chapter is occasionally marred by rhetoric ('... many older ideas [i.e. secondary structure of proteins] can now be seen to be naïve'; 'confusing biology with chemistry'), but it is a vigorously written and highly readable manifesto, and would still be recommended reading if its only virtue were the impressive breadth of protein physico-chemistry on which it draws.

This is not a book for the generalist; it presupposes fluency in thermodynamics, kinetics and spectroscopy, and the reader may occasionally inkle that Hilbert's dictum 'physics has become too difficult for physicists' is beginning to embrace biochemistry. However, the contributions, diverse as they are, are uniformly well written (this may the result of careful editorial work), and the collection will be of value to anyone with an active interest in the physical chemistry of proteins.

Paul Woolley