MOLECULAR BIOLOGY OF THE CELL

Bruce Alberts • Dennis Bray
Julian Lewis • Martin Raff • Keith Roberts
James D. Watson

Garland Publishing, Inc.
New York & London

"Long ago it became evident that the key to every biological problem must finally be sought in the cell, for every living organism is, or at sometime has been, a cell."

Edmund B. Wilson
The Cell in Development and Heredity
3rd edition, 1925, Macmillan, Inc.

Bruce Alberts received his Ph.D. from Harvard University and is currently a Professor in the Department of Biochemistry and Biophysics at the University of California Medical School in San Francisco. *Dennis Bray* received his Ph.D. from the Massachusetts Institute of Technology and is currently a Senior Scientist in the Medical Research Council Cell Biophysics Unit at King's College London. *Julian Lewis* received his D.Phil. from Oxford University and is currently a Lecturer in the Anatomy Department at King's College London. *Martin Raff* received his M.D. degree from McGill University and is currently a Professor in the Zoology Department at University College London. *Keith Roberts* received his Ph.D. from Cambridge University and is currently Head of the Department of Cell Biology at the John Innes Institute, Norwich. *James D. Watson* received his Ph.D. from the University of Indiana and is currently Director of the Cold Spring Harbor Laboratory. He is the author of *Molecular Biology of the Gene* and, with Francis Crick and Maurice Wilkins, won the Nobel Prize in Medicine and Physiology in 1962.

Library of Congress Cataloging in Publication Data
Main entry under title:

Molecular biology of the cell.

Includes bibliographies and index.
1. Cytology. 2. Molecular biology. I. Alberts, Bruce, 1938– [DNLM: 1. Cells. 2. Molecular biology. QH 581.2 M718]
QH581.2.M64 1983 574.87 82-15692
ISBN 0-8240-7282-0

Published by Garland Publishing, Inc.
136 Madison Avenue, New York, NY 10016

Printed in the United States of America

15 14 13 12 11 10 9 8 7 6 5 4 3

Preface

There is a paradox in the growth of scientific knowledge. As information accumulates in ever more intimidating quantities, disconnected facts and impenetrable mysteries give way to rational explanations, and simplicity emerges from chaos. Gradually the essential principles of a subject come into focus. This is true of cell biology today. New techniques of analysis at the molecular level are revealing an astonishing elegance and economy in the living cell and a gratifying unity in the principles by which cells function. This book is concerned with those principles. It is not an encyclopedia but a guide to understanding. Admittedly, there are still large areas of ignorance in cell biology and many facts that cannot yet be explained. But these unsolved problems provide much of the excitement, and we have tried to point them out in a way that will stimulate readers to join in the enterprise of discovery. Thus, rather than simply present disjointed facts in areas that are poorly understood, we have often ventured hypotheses for the reader to consider and, we hope, to criticize.

Molecular Biology of the Cell is chiefly concerned with eucaryotic cells, as opposed to bacteria, and its title reflects the prime importance of the insights that have come from the molecular approach. Part I and Part II of the book analyze cells from this perspective and cover the traditional material of cell biology courses. But molecular biology by itself is not enough. The eucaryotic cells that form multicellular animals and plants are social organisms to an extreme degree: they live by cooperation and specialization. To understand how they function, one must study the ways of cells in multicellular communities, as well as the internal workings of cells in isolation. These are two very different levels of investigation, but each depends on the other for focus and direction. We have therefore devoted Part III of the book to the behavior of cells in multicellular animals and plants. Thus developmental biology, histology, immunobiology, and neurobiology are discussed at much greater length than in other cell biology textbooks. While this material may be omitted from a basic cell biology course, serving as optional or supplementary reading, it represents an essential part of our knowledge about cells and should be especially useful to those who decide to continue with biological or medical studies. The broad coverage expresses our conviction that cell biology should be at the center of a modern biological education.

This book is principally for students taking a first course in cell biology, be they undergraduates, graduate students, or medical students. Although we assume that most readers have had at least an introductory biology course, we have attempted to write the book so that even a stranger to biology could follow it by starting at the beginning. On the other hand, we hope that it will also be useful to working scientists in search of a guide to help them pick their way through a vast field of knowledge. For this reason, we have provided a much more thorough list of references than the average undergraduate is likely to require, at the same time making an effort to select mainly those that should be available in most libraries.

This is a large book, and it has been a long time in gestation—three times longer than an elephant, five times longer than a whale. Many people have had a hand in it. Each chapter has been passed back and forth between the author who wrote the first draft and the other authors for criticism and revision, so that each chapter represents a joint composition. In addition, a small number of outside experts contributed written material, which the authors reworked to fit with the rest of the book, and all the chapters were read by experts, whose comments and corrections were invaluable. A full list of acknowledgments to these contributors and readers for their help with specific chapters is appended. Paul R. Burton (University of Kansas), Douglas Chandler (Arizona State University), Ursula Goodenough (Washington University), Robert E. Pollack (Columbia University), Robert E. Savage (Swarthmore College), and Charles F. Yocum (University of Michigan) read through all or some of the manuscript and made many helpful suggestions. The manuscript was also read by undergraduate students, who helped to identify passages that were obscure or difficult.

Most of the advice obtained from students and outside experts was collated and digested by Miranda Robertson. By insisting that every page be lucid and coherent, and by rewriting many of those that were not, she has played a major part in the creation of a textbook that undergraduates will read with ease. Lydia Malim drew many of the figures for Chapters 15 and 16, and a large number of scientists very generously provided us with photographs: their names are given in the figure credits. To our families, colleagues, and students we offer thanks for forbearance and apologies for several years of imposition and neglect. Finally, we owe a special debt of gratitude to our editors and publisher. Tony Adams played a large part in improving the clarity of the exposition, and Ruth Adams, with a degree of good-humored efficiency that put the authors to shame, organized the entire production of the book. Gavin Borden undertook to publish it, and his generosity and hospitality throughout have made the enterprise of writing a pleasure as well as an education for us.

We welcome readers' suggestions and corrections, which should be sent to us c/o Gavin Borden, Garland Publishing, Inc., 136 Madison Avenue, New York, NY 10016.

Acknowledgments

Chapter 1
Read in whole or in part by: Tom Cavalier-Smith (King's College London), Monroe Strickberger (University of Missouri, St. Louis).

Chapter 2
Read in whole or in part by: Charles Gilvarg (Princeton University), Phillips Robbins (Massachusetts Institute of Technology).

Chapter 3
Read in whole or in part by: Robert Fletterick (University of California, San Francisco), Phillips Robbins (Massachusetts Institute of Technology).

Chapter 4
Read in whole or in part by: David Sabatini (New York University School of Medicine), John Sedat (University of California, San Francisco), Patrick O'Farrell (University of California, San Francisco), Fred Richards (Yale University).

Chapter 5
Read in whole or in part by: Glenn Herrick (University of Utah), Patrick O'Farrell (University of California, San Francisco), Brian McCarthy (University of California, Irvine).

Chapter 6
Read in whole or in part by: Mark Bretscher (MRC Laboratory of Molecular Biology, Cambridge, England), Bastien Gomperts (University College Hospital Medical School, London), Juan Korenbrot (University of California, San Francisco), Dale Oxender (University of Michigan).

Chapter 7
Major contribution: James Rothman (Stanford University).

Read in whole or in part by: George Palade (Yale University), Daniel Friend (University of California, San Francisco), Gary Firestone (University of California, Berkeley).

Chapter 8
Read in whole or in part by: Michael Ashburner (Cambridge University), Pierre Chambon (University of Strasbourg), Patrick O'Farrell (University of California, San Francisco), Joseph Gall (Yale University), Larry Gerace (Johns Hopkins University), Glenn Herrick (University of Utah), E. G. Jordan (Queen Elizabeth College, London), Brian McCarthy (University of California, Irvine), Robert Perry (Institute of Cancer Research, Philadelphia), Abraham Worcel (University of Rochester).

Chapter 9
Major contribution: Peter Garland (University of Dundee, Scotland), Piet Borst (Jan Swammerdam Institute, University of Amsterdam).

Read in whole or in part by: Martin Brand (Cambridge University), Roderick Capaldi (University of Oregon), Richard McCarty (Cornell University),

Allison Smith (John Innes Institute, Norwich, England), Charles Yocum (University of Michigan).

Chapter 10
Additional material: Marc Kirschner (University of California, San Francisco).

Read in whole or in part by: Roger Cooke (University of California, San Francisco), Graham Dunn (MRC Cell Biophysics Unit, London), John Kendrick-Jones (MRC Laboratory of Molecular Biology, Cambridge, England), Marc Kirschner (University of California, San Francisco), Klaus Weber (Max Planck Institute for Biophysical Chemistry, Göttingen).

Chapter 11
Additional material: J. Michael Bishop (University of California, San Francisco), Jeremy Pickett-Heaps (University of Colorado).

Read in whole or in part by: Leland Hartwell (University of Washington), J. Murdoch Mitchison (University of Edinburgh), Zacheus Cande (University of California, Berkeley), John Wyke (Imperial Cancer Research Fund, London).

Chapter 12
Additional material: Günther Gerisch (Max Planck Institute for Biochemistry, Martinsried), Robert Trelstad (Rutgers Medical School).

Read in whole or in part by: Stephen Burden (Harvard Medical School), Max Burger (University of Basel), Bernie Gilula (Baylor University), Zach Hall (University of California, San Francisco), Richard Hynes (Massachusetts Institute of Technology), Colin Manoil (Harvard Medical School), Darwin Prockop (Rutgers Medical School), David Rees (National Institute for Medical Research, Mill Hill, London), John Scott (University of Manchester), Malcolm Steinberg (Princeton University).

Chapter 13
Additional material: Michael Wilcox (MRC Laboratory of Molecular Biology, Cambridge, England).

Read in whole or in part by: Peter Baker (King's College London), Philip Cohen (University of Dundee), Daniel Koshland (University of California, Berkeley), Anne Mudge (University College London), Jesse Roth (National Institutes of Health, Bethesda), Charles Stevens (Yale University).

Chapter 14
Major contribution: David Epel (Stanford University)

Read in whole or in part by: Adelaide Carpenter (University of California, San Diego), Jeffrey Hall (Brandeis University), Anne McLaren (University College London), Montrose Moses (Duke University), Duncan O'Dell (University College London), David Phillips (Rockefeller University), Lewis Tilney (University of Pennsylvania).

Chapter 15
Major contribution: Cheryll Tickle (Middlesex Hospital Medical School, London).

Additional material: Judith Kimble (University of Wisconsin).

Read in whole or in part by: John Gerhart (University of California, Berkeley), Peter Lawrence (MRC Laboratory of Molecular Biology, Cambridge, England), Anne McLaren (University College London), Norman Wessells (Stanford University), Lewis Wolpert (Middlesex Hospital Medical School, London).

Chapter 16
Additional material: Vernon Thornton (King's College London).

Read in whole or in part by: Barry Brown (King's College London), Judah

Folkman (Harvard Medical School), Peter Gould (Middlesex Hospital Medical School, London), Jay Lash (University of Pennsylvania), John Owen (University of Birmingham, England), Philippe Sengel (University of Grenoble), Cheryll Tickle (Middlesex Hospital Medical School, London), Rosalind Zalin (University College London).

Chapter 17
Read in whole or in part by: Leroy Hood (California Institute of Technology), Peter Lachmann (MRC Center, Cambridge, England), Avrion Mitchison (University College London), Alan Munro (Cambridge University), Hans Müller-Eberhard (Scripps Clinic and Research Institute), William Paul (National Institutes of Health, Bethesda), Robert Schreiber (Scripps Clinic and Research Institute), Martin Weigert (Institute of Cancer Research, Philadelphia).

Chapter 18
Major contribution: Charles Stevens (Yale University).

Additional material: Regis Kelly (University of California, San Francisco).

Read in whole or in part by: Jonathan Ashmore (University of Sussex, England), Peter Baker (King's College London), Darwin Berg (University of California, San Diego), Anne Mudge (University College London).

Chapter 19
Major contribution: Brian Gunning (Australian National University, Canberra).

Read in whole or in part by: Jim Dunwell (John Innes Institute, Norwich, England), Ray Evert (University of Wisconsin, Madison), Larry Fowke (University of Saskatchewan, Saskatoon), John Hall (University of Southampton, England), David Hanke (University of Cambridge, England), Andy Johnston (John Innes Institute, Norwich, England), Virginia Walbot (Stanford University), Trevor Wang (John Innes Institute, Norwich, England), John Watts (John Innes Institute, Norwich, England).

Cover photograph kindly provided by Michael Verderame and Robert Pollack of Columbia University. The fluorescein-phalloidin used to stain the actin cables was the generous gift of Drs. Theodore Wieland and A. Deboben of the Max Planck Institute, West Germany. The photograph is of a mouse fibroblast that had been transformed to anchorage-independent growth by the virus Simian Virus 40 (SV40) and subsequently selected for anchorage-dependent growth. This particular cell was stained for SV40 large T antigen (*red*) and fluorescein-phalloidin (*green*), which specifically stains F actin.

Prologue

It is all too easy now to underestimate cells. We have known about them for such large fractions of our lives that, for the most part, we cease being aware of how remarkable they really are. Almost as soon as we learn our first rudiments of science, we are told that all living beings are formed from cells, that cells come into existence from the growth and division of preexisting ones, and that they may exist either singly as unicellular organisms or as parts of immensely complicated organisms that may contain billions of interacting, highly specialized units. It has been their potential for great diversity of size, shape, and function that has allowed evolution to proceed in such strikingly different directions.

The mere cataloging of the different names and unique properties of cells has very limited intellectual appeal. But the textbook dry facts take on new meaning when we first use our simple school microscopes to look at the tiny one-celled creatures like the amoebae or paramecia that inhabit drops of pond water. Then the cell as an amazing moving body comes alive, and it is natural to wonder what exact molecules it is made of and how it can so regularly grow and divide to provide more of its kind. Until the 1950s, however, this objective seemed far beyond our capabilities as scientists. Up to then we had little choice but to focus upon the descriptive morphological approach, using better and better microscopes to reveal more and more cellular structures. To these we frequently gave fancy names, like the ergastoplasm or chondriosomes, without understanding why they were there or how they functioned. Not surprisingly, many found this approach unsatisfying and moved on from cells per se to explore the underlying chemical reactions that were becoming increasingly amenable to logical analysis.

These "biologists turned biochemists" soon discovered how cells use the energy in food molecules to build up new biological molecules, thereby discovering how cells can grow and divide without disobeying the thermodynamic dictum that all chemical reactions must move in the direction that maximizes the production of heat and disorder. This momentous achievement greatly encouraged the increasing number of scientists who thought the essence of cells lay entirely in their molecular organization and the enzymatically mediated pathways by which their molecules are either broken down or built up. There was, however, still great uncertainty about where genes fitted into the chemical picture, and particularly whether they had a direct role in correctly linking together the hundreds of amino acids that make a typical protein molecule. How this might occur was still conceptually quite unclear as late as the 1940s, and no one expected the incredibly rapid pace at which the nature and transmission of genetic information was worked out between 1953 and 1966. Then with the dominant role of DNA so clearly established, it was very tempting to say that by understanding the nucleic acids we had understood the essence of the living state and that the greatest challenges of biology had been surmounted.

This is a view that we do not share. The interconnecting pathways between ATP and DNA, marvelous as they are, do not give us the living cell. Even the simplest cells are far more complex objects than generally perceived and vastly more ingenious than any computerized intelligent machine yet designed. That this is so is strongly hinted at by simple observation. We do not have to see through the outer surfaces of cells to appreciate that the biological organization that permits them to act in such rational ways must indeed be incredibly subtle and versatile.

Consider for example the extraordinarily complex changes in cellular shape that accompany the movements of fibroblasts. These connective tissue cells are the principal makers of the extracellular matrix that helps glue together the tissues of multicellular organisms. Within animals, fibroblasts must constantly be prepared to move into areas of newly forming tissue. Removed from the animal and grown in culture, they are accessible to microscopic analysis, and the morphological changes that accompany their passage from one point to another have been extensively documented through cinematographic analysis.

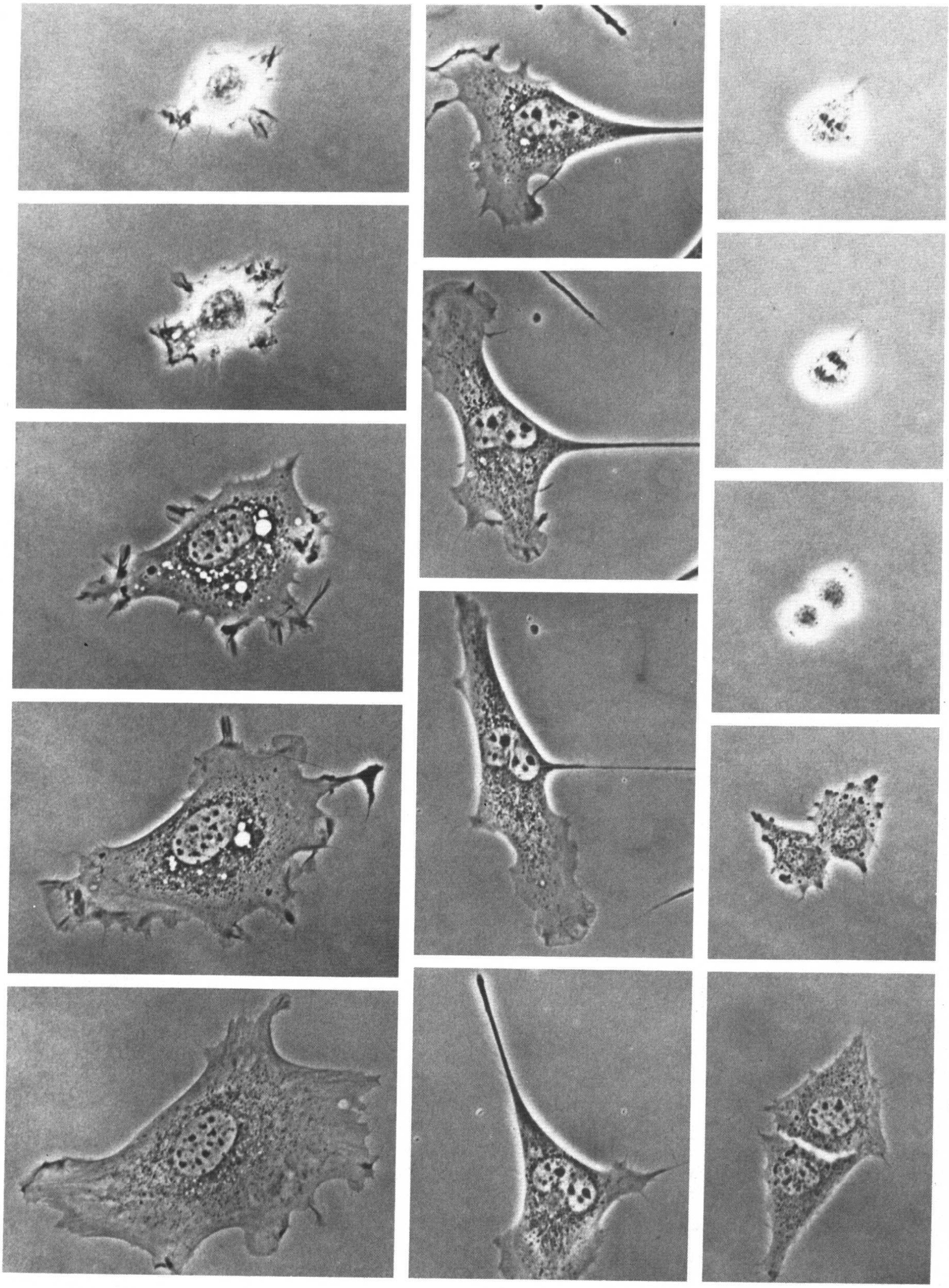
The isolated well-fed single fibroblast is thus revealed to be a restless, apparently unsatisfied creature unable to stay quiet but instead internally programmed to move. It and all its progeny will continue moving and growing until the flat surface of the plastic dish is covered with a single layer of closely packed cells. Moving fibroblasts bear a resemblance to amoebae with their extending pseudopodia (false feet) that we first saw in our early school science classes, but the details of their respective motions are not the same.

Fibroblast movement is initiated by the rapid, virtually frenetic, throwing out of filamentous extensions (microspikes) and sheetlike projections (lamellipodia). Each of these projections can make firm attachments to the underlying surfaces ahead. Such attachments lead to a forward flow of the cell's cytoplasm and its enclosed nucleus. Many many more such locomotor protuberances are pushed out than ever make firm attachments, and those that fail to attach are swept up as "ruffles" in the backward flow of the upper cell surface that eventually sends them to the rear of the cell. Large numbers of potential adhesion points can thus be sampled, with firm union made only to the most favorable sites.

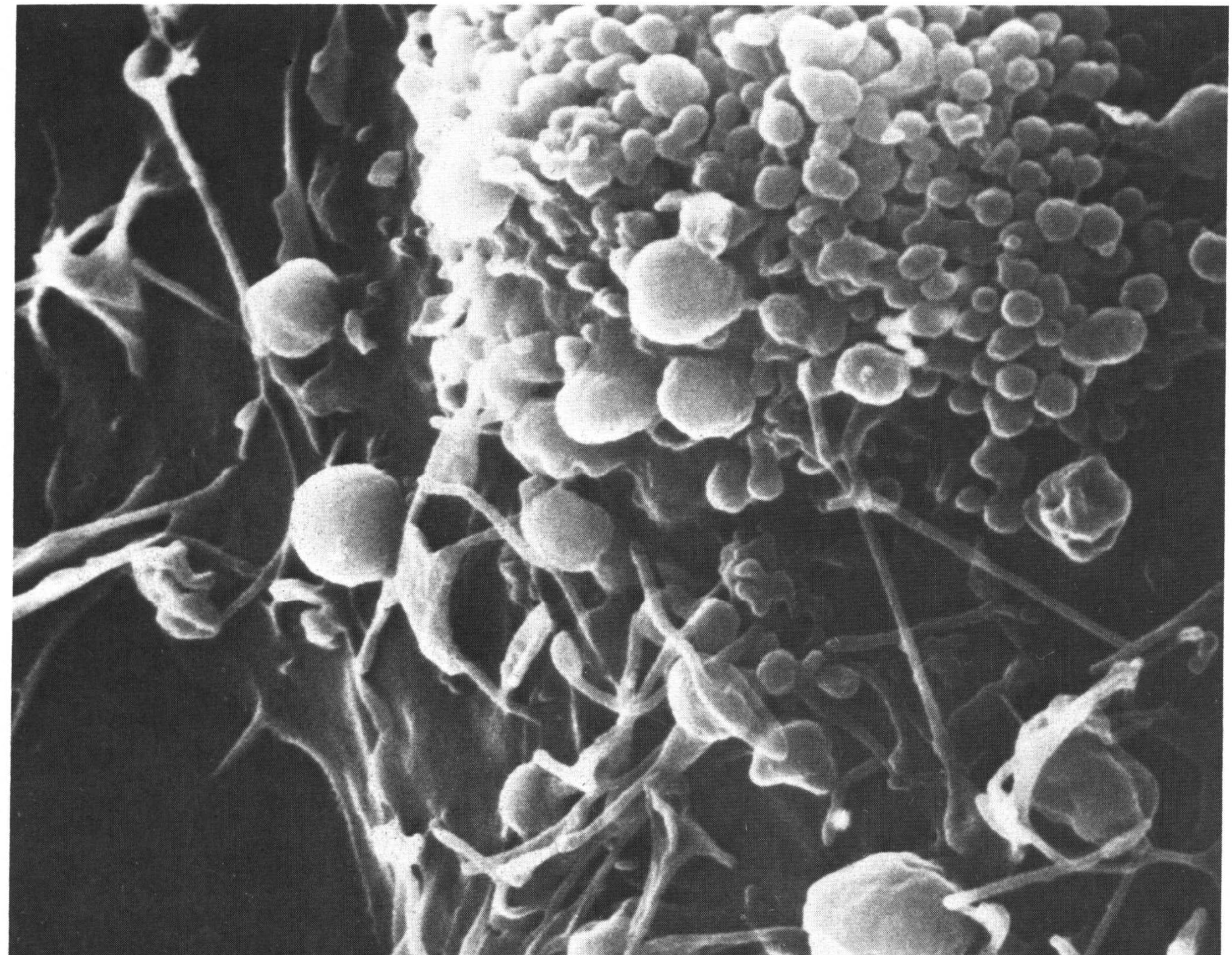
The capacity of fibroblasts for long persistent solo excursions is not a property of all cells. When, for example, single epithelial cells of the sort that line our intestines or skin are placed in culture, they show no tendency to move about. The locomotor behavior of a given cell type thus appears to be highly foreordained and, like virtually all other cellular events of consequence, is hardly ever left to chance. As a result, the exact final position of a given cell within a multicellular organism arises from a myriad of well-regulated biochemical steps that effectively give the cell no choice but to come together harmoniously with other cells in a particular configuration.

To understand how these logical steps unfold, we clearly must probe beneath the cell surface. Happily we now possess the highly sophisticated microscopic, biochemical, and genetic engineering procedures to let us tackle on virtually equal odds the cell's almost overwhelming complexity. We have already found that the apparently amorphous cytoplasmic mass contains interlaced patterns of specific fibrous protein aggregates. These filamentous structures, themselves built of smaller subunit protein molecules, are assembled into the elaborate scaffolds and molecular machines that give rise to directed cell movements.

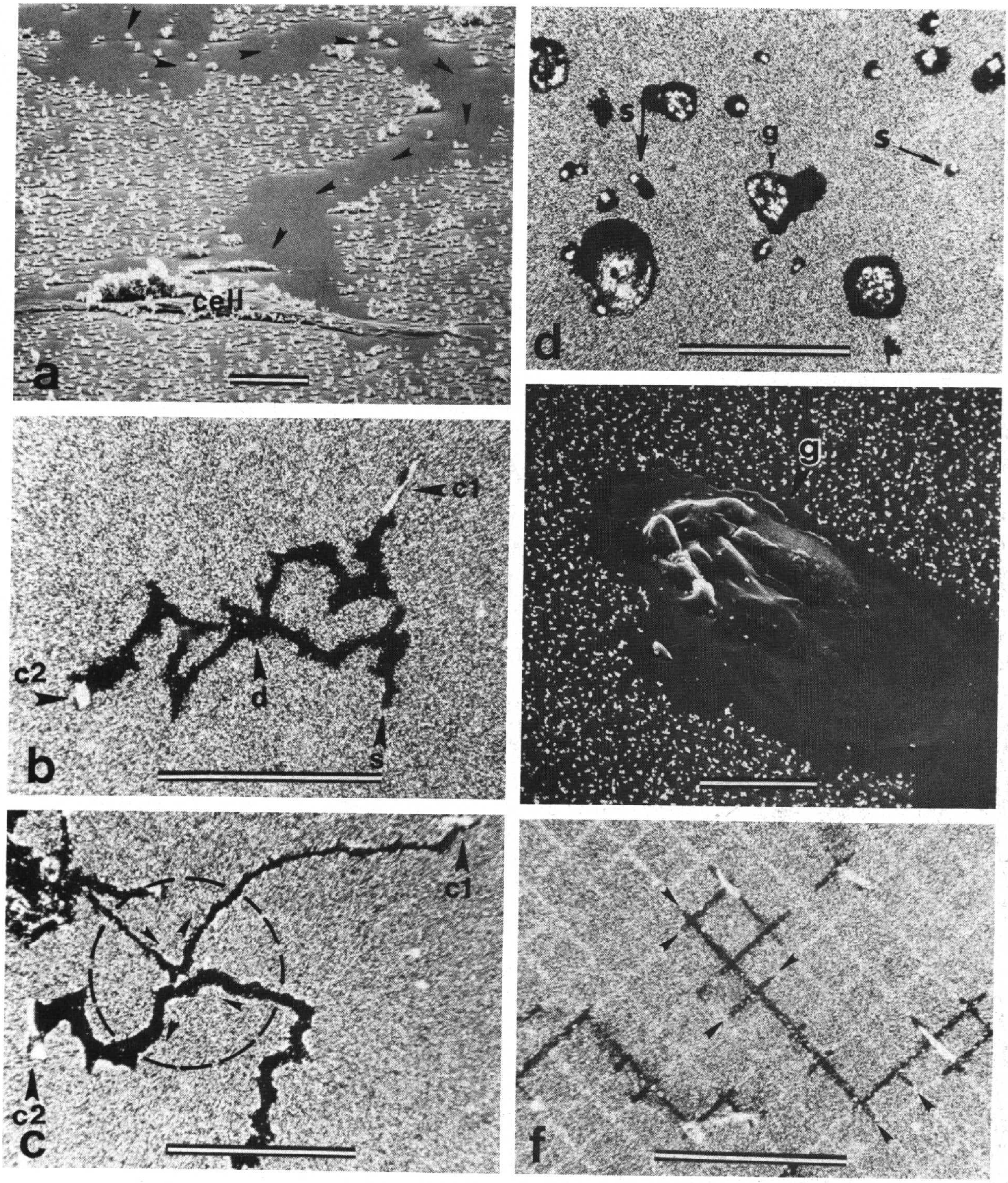
As this book unfolds, we shall relate how various cell structural elements are built and maintained by specific interactions between complex molecules. And with less precision we shall outline how they enable a cell to grow and divide and how they generate the metamorphoses of the cell's architecture that we call cell movement and differentiation, which enable cells to participate in the construction of multicellular organisms. We hope that we shall also convey the sense of great mystery that surrounds the many problems that we do not yet know quite how to handle, the feeling of marvelous excitement that comes from the great achievements of today's cell biology, and, last but not least, the logical as well as the optical beauty of cells.



Mobile behavior of mouse fibroblasts (3T3 cells), as revealed by phase-contrast microscopy. (*Left panel*) As the cell flattens down upon a surface, needlelike microspikes and sheetlike lamellipodia are projected outward to seek suitable attachment sites. Intermittently, the lamellipodia fold back on themselves ("ruffle") before extending again. (*Center panel*) After flattening down, fibroblasts assume a polarized shape with leading lamellipodia and begin to crawl along the surface of the culture dish. In this series of four micrographs we observe an abrupt change in direction. (*Right panel*) In going through mitosis, a flattened cell rounds up prior to formation of the mitotic spindle; the two daughter cells reassume a flattened position following their separation. In each panel, successive micrographs taken at successive times are displayed from top to bottom. (Courtesy of Guenter Albrecht-Buehler.)

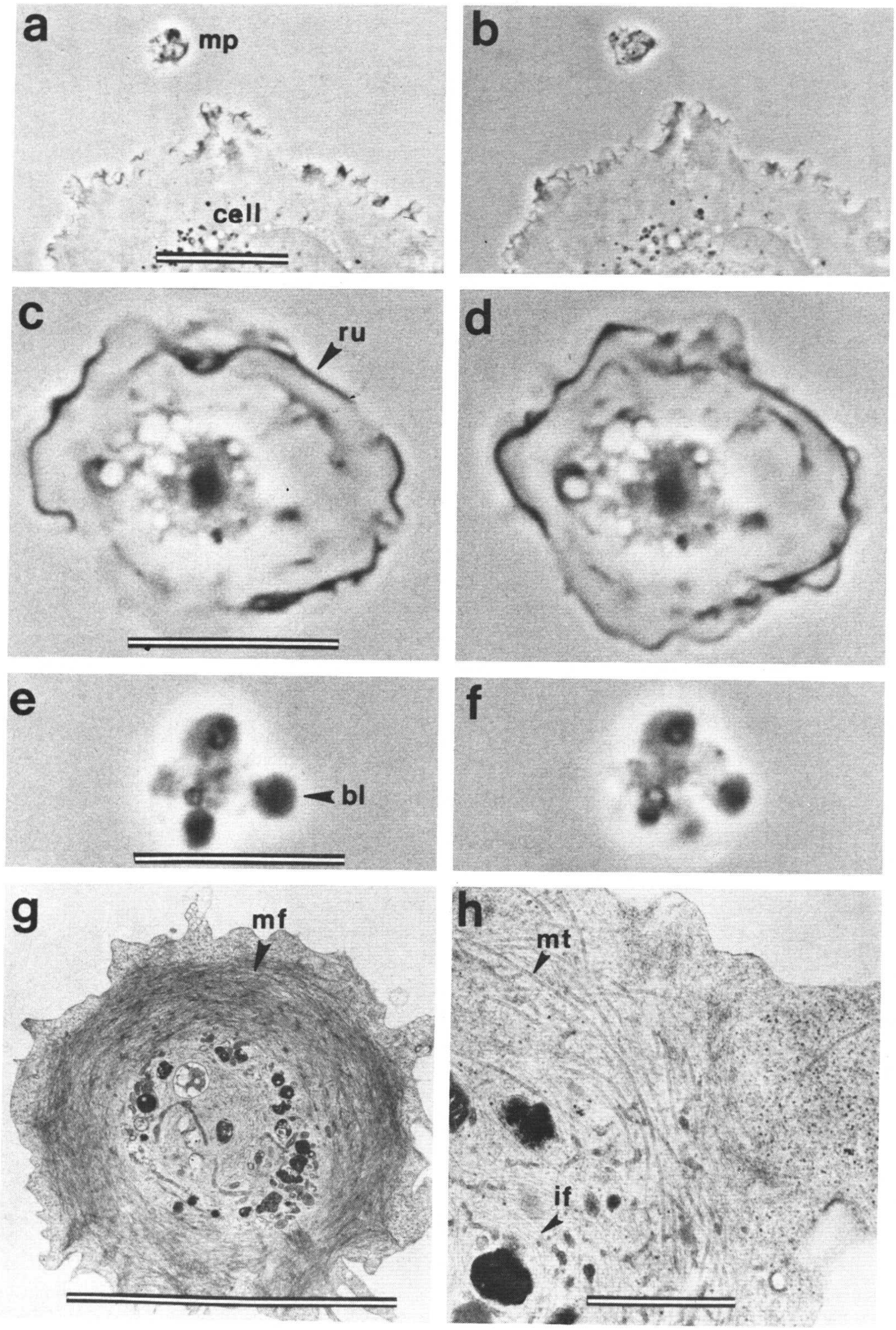


A scanning electron microscopic view of part of the surface of a mouse fibroblast in the process of flattening. Microspikes and lamellipodia project outward at the ruffling edges while large numbers of hemispherelike projections ("blebs") project from the region over the centrally located nucleus. The insert shows a lower-magnification view of the entire cell, whose diameter is about 25 μm. (Courtesy of Guenter Albrecht-Buehler.)



Paths of cell migration are revealed by cells moving on a surface coated with tiny gold particles. Extending microspikes and lamellipodia pick up the loose gold particles and help bring them back over the cell body where they are engulfed (phagocytosed) into the cell. Such events create clearings in regions through which cells have moved. In dark-field illumination, the tracks appear black while the gold-particle-filled cells glow brightly. (Courtesy of Guenter Albrecht-Buehler; reproduced from *J. Natl. Cancer Inst.* Monograph 60, 1982.)

(a) Scanning electron micrograph of a track, showing the many tiny gold particles on the substrate, the cell, and the particle-free track left behind by the cell (*arrows*). Bar indicates 20 μm. (b) Dark-field micrograph of the branching track of a mother 3T3 mouse cell that started at "s" and divided at "d" into the two similar tracks of the sister cells "c1" and "c2." Bar indicates 500 μm. (c) Collision between two 3T3 mouse cells (c1 and c2). Within the circled area the two cells have bounced off each other as if they were colliding billiard balls. Bar indicates 500 μm. (d, e) Dark-field light micrograph (d) and scanning electron micrograph (e) of migrating PTK 1 (rat kangaroo) cell groups (g). In (d), "s" points to single cells that migrated little. Bars indicate 500 μm (d) and 50 μm (e). (f) Tracks of guided 3T3 cells (*bright white structures*) on a checkerboard of guiding lines (*whitish lines*). The cells follow the lines but at the intersections probe into optional directions, as indicated by the small sideways "thorns" in the tracks at these points (arrowheads point to a few of these thorns). Bar indicates 500 μm.



Very small autonomously moving cellular fragments (microplasts) are often generated following exposure of cells to the cytoskeleton-disrupting drug cytochalasin B. Though they lack a nucleus, microplasts can flatten, ruffle, and bleb, showing their possession of organized functional cytoskeletal elements. (Courtesy of Guenter Albrecht-Buehler; reproduced from *J. Natl. Cancer Inst.* Monograph 60, 1982.)

(a, b) A ruffling microplast (mp) near the edge of a flattening human cell (cell) for size comparison. Bar indicates 20 μm. Time lapse between the two pictures is 35 seconds, showing the movement. (c, d) Two photographs of a ruffling microplast taken 15 seconds apart. One of the many ruffles is indicated by "ru." Bar indicates 10 μm. (e, f) Two photographs of a blebbing microplast taken 10 seconds apart. One of the many blebs is indicated by "bl." Bar indicates 10 μm. (g, h) Electron micrographs of a typical microplast sectioned parallel to the flat surface on which it sits. Visible are peripheral actin-containing microfilaments (mf), microtubules (mt), and intermediate filaments (if). Bars indicate 10 μm (g) and 1 μm (h).

Contents in Brief

List of Topics

PART I
Introduction to the Cell

CHAPTER 1
The Evolution of the Cell

CHAPTER 2
Small Molecules, Energy, and Biosynthesis

CHAPTER 3

Macromolecules: Structure, Shape, and Information