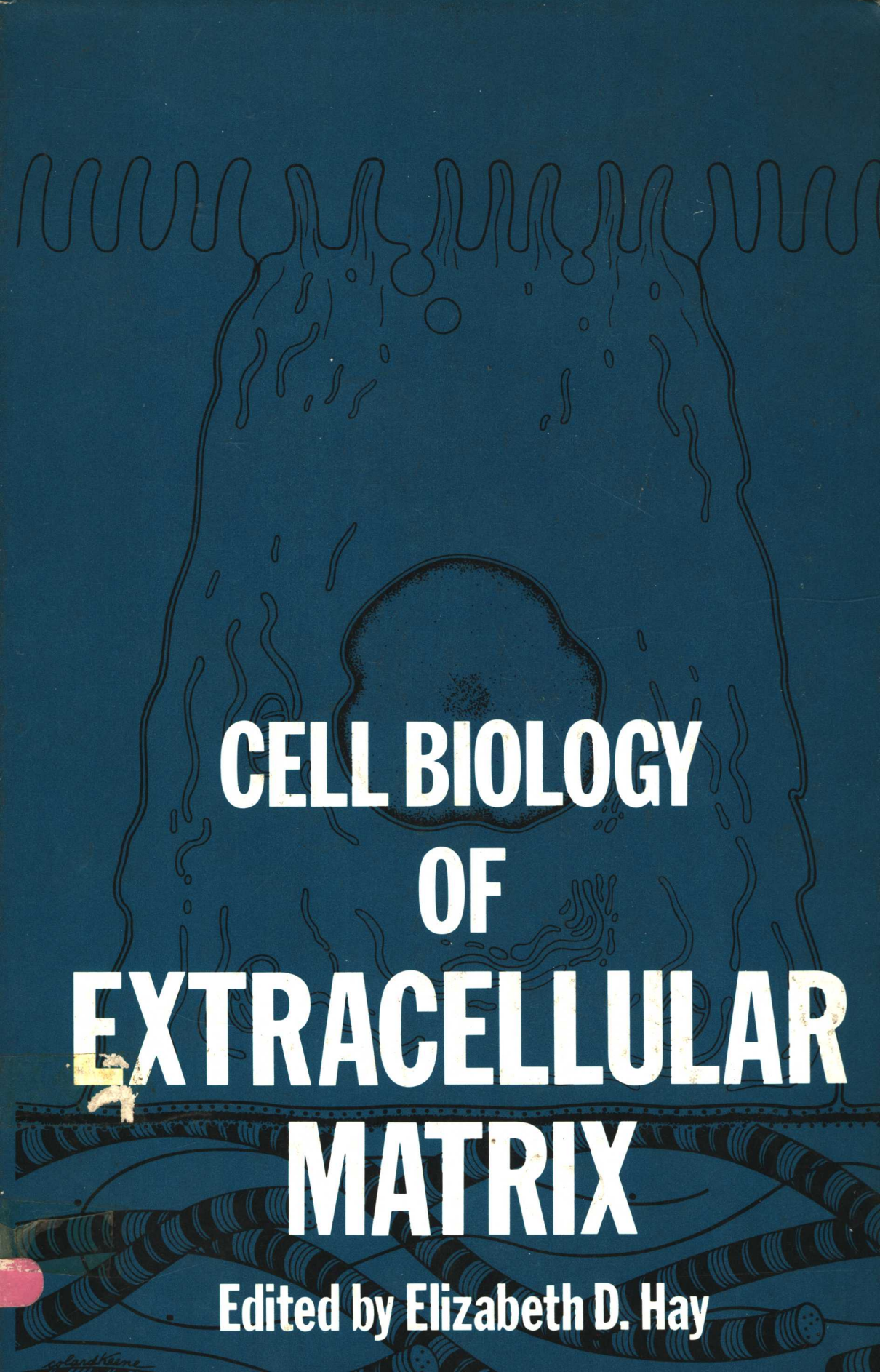

# CELL BIOLOGY OF EXTRACELLULAR MATRIX

Edited by Elizabeth D. Hay

# Cell Biology of Extracellular Matrix

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# Preface

At a recent meeting to discuss the domains of cell biology, I put forth a case for the extracellular matrix, even though my argument ran the risk of falling on deaf ears. After all, the matrix is EXTRAcellular, outside the cells. In this book, however, the authors make a compelling case for the relevance of the matrix to cellular concerns. Not only are numerous cell types, including many epithelia, quite caught up in the business of manufacturing matrix components, but also most of them contain matrix molecules in exoskeletons that are attached to the plasmalemma and that organize or otherwise influence the affairs of the cytoplasm. The idea of this book is to present the extracellular matrix to cell biologists of all levels. The authors are active and busy investigators, recognized experts in their fields, but all were enthusiastic about the prospect of writing for this audience. The chapters are not "review" articles in the usual sense, nor are they rehashes of symposium talks; they were written specifically for this book and they present the "state of the art" in engaging style, with ample references to more technical or historical reviews. The book is rich in electron micrographs and diagrams and for many of the latter, as well as for the design of the cover, we are indebted to Sylvia J. Keene, medical illustrator for the Department of Anatomy at Harvard Medical School. We also owe special thanks to Susan G. Hunt of this Department, who did a masterful preliminary job of editing the manuscripts, and to the editors and staff of Plenum Press, who were supporting and helpful throughout the enterprise. We think you will enjoy this treatise.

Elizabeth D. Hay

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# Introductory Remarks

ELIZABETH D. HAY

Cytoskeleton, cell shape, cell migration, control of cell growth and differentiation, these are all subjects that to be fully understood today require a consideration of the extracellular matrix (ECM): its composition, its role in development, and its relation to the cell surface. The ECM is the structurally stable material that lies under epithelia and surrounds connective tissue cells, but the old concept of the ECM as an inert supporting material, created by the cells as a mere scaffolding on or in which to reside, is now bygone. Surely, collagens are sources of strength to the tissues, elastin and proteoglycans are essential to matrix resiliency, and the structural glycoproteins help to create tissue cohesiveness. But the cell, having produced these extracellular macromolecules, and spoken out in one way or another on the question of their assembly, does not then divorce itself of them. The cell continues to interact with its own ECM products, and with the ECM produced by other cells. At the cell surface, a structural and functional continuum seemingly is formed between the cell interior, the cell membrane, and the molecules of the matrix, so that the metabolism and fate of the cell, its shape, and many of its other properties are continuously related to and dependent on the composition and organization of the matrix.

The ECM is composed of a far greater variety of molecules than we would have guessed even a decade ago. Discounting all the molecules that must pass through the matrix to reach the cells or are temporarily trapped in the ECM, we are left with a basic structural composition of at least four major classes of macromolecules. In Chapter 1, Dr. Linsenmayer brings us up to date on the chemistry and molecular biology of the five or more different (genetically distinct) types of collagen molecules. Then (Chapter 2), Drs. Vincent and Gretchen Hascall describe the remarkable progress that has been made in recent years in our knowledge of proteoglycan composition and assembly. In Chapter 3, Drs. Franzblau and Faris describe a unique protein of the ECM, elastin, and give us insights into its ultrastructure and the development of elastic fibers. The final class of ECM molecules, the structural glycoproteins, are relatively large molecules with sugar side chains that are longer

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than those found in collagen and are often rich in sialic acid. The structural glycoproteins are discussed in Chapter 4 by Dr. Yamada, who has pioneered many of the recent discoveries on the structure and function of fibronectin.

A book on the cell biology of extracellular matrix would, of course, consider the manner in which cells synthesize ECM, influence its organization, and bring about its degradation. These subjects are covered in the second section of the book. In Chapter 5, Dr. Dorfman describes the biosynthesis of glycosaminoglycans (GAG) and the protein cores that, together with GAG, form the proteoglycan (PG) monomers that become assembled into the PG aggregates of the ECM; in both Chapters 2 and 5, the authors take care to relate biochemical aspects of PG organization and synthesis to tissue and cell ultrastructure. The role of the endoplasmic reticulum and Golgi complex in the synthesis and secretion of collagens is developed in Chapter 6 by Dr. Olsen, who also informs us of the very recent and exciting advances that have been made in the area of collagen gene structure and function. Drs. Trelstad and Silver (Chapter 7) present a very original synthesis of our understanding of the physical chemistry, molecular biology, and morphology of collagen assembly. Finally (Chapter 8), Dr. Gross acquaints us with what is known and not known about collagenases and the process of ECM degradation (and remodeling) that somehow, in amazing fashion, is controlled by the cells.

The matrix, as we indicated earlier, is, in a sense, talking back to the cells that create (and reside on or in) its interstices. In the third section of the book, we consider the phenomenon of cell–extracellular matrix communication in more detail. Dr. Toole (Chapter 9) tells us about the role of hyaluronic acid and PG in morphogenesis, cell migration, and cell proliferation, and he presents a model of interaction of hyaluronic acid with the cell surface. Dr. Hynes (Chapter 10) then presents a model of interaction of fibronectin with other ECM molecules and with the cell surface (the models are not necessarily coexclusive). Chapter 10 also reviews *in vitro* studies of the role of fibronectin in cell growth, cell migration, platelet adhesion, etc., and speculates on its possible *in vivo* functions. In Chapter 11, Dr. Farquhar convinces us that the PG component of the glomerular basement membrane (GBM) plays an important role in the filtering function of the glomerulus, and in the last chapter, I give an overview of the effects of collagens on cell differentiation, cell shape, and cell metabolism, and speculate further on the relation of the ECM to the cell surface.

We are fortunate in being able to bring together so many investigators who are pioneers in their fields and were so willing to make the advances in, and future of, these important subjects clear to the cell biologist. In editing this endeavor, I have not attempted to impose my own views of terminology on these distinguished authors. Thus, the reader will find that some terms are used interchangeably, the best example being the terms *basal lamina* and *basement membrane*. The basal (basement) lamina or basement membrane is a zone about 100 nm wide under epithelia and around muscle cells; it consists of a central compact sheet of collagen (and probably other glycoproteins), called the lamina densa, that is separated from the cell by a less electron-dense zone, the lamina rara externa, and from the underlying connective tissue by a second electron-

lucid zone, the lamina rara interna. Both the laminae rarae externa and interna contain a layer of PG granules connected by small filaments to the cell, on the one hand, and to underlying collagen fibrils, on the other; these structures are not visible unless special fixatives are used, hence the term *rara* (or *lucida*) is used to refer to the "empty" zone (Fig. i-1).

The term *basement membrane* was originally used by light microscopists to refer to the whole condensation of connective tissue (basal lamina and the

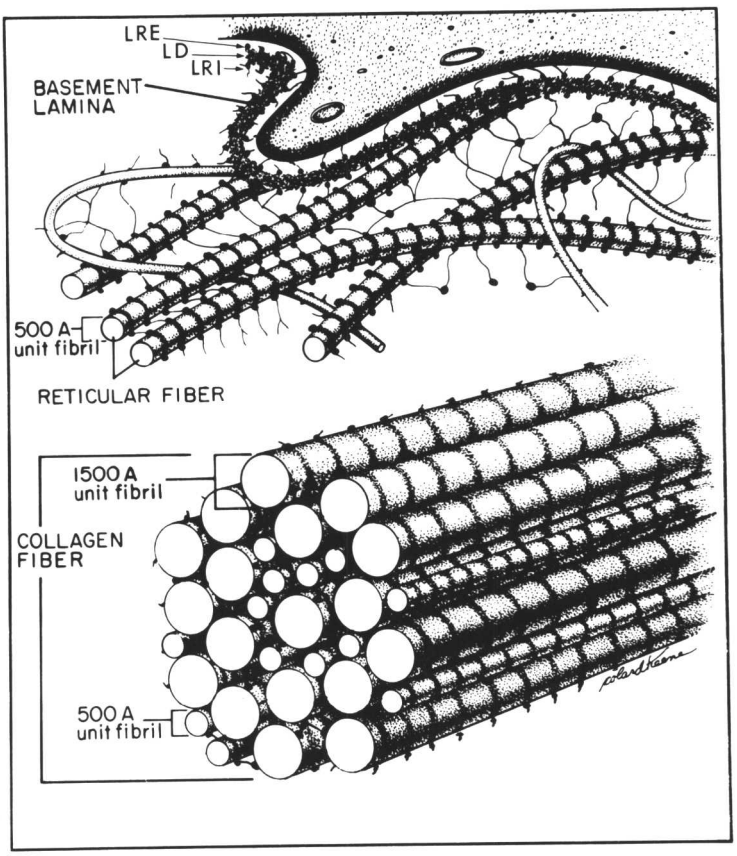


**Figure i-1.** Diagrams depicting relations of proteoglycans, collagen fibrils, and basement lamina in the tissues. The drawing at the top includes part of the basal cytoplasm of an epithelial cell. The outermost layer of the basal lamina, the lamina rara externa (LRE), is attached to the epithelial cell by small filaments presumably composed of glycoprotein. The LRE and LRI (lamina rara interna) each contain a layer of proteoglycan granules. The lamina densa (LD) contains collagen and possibly other glycoproteins (see Chapter 11). Collagen fibrils associated with the basement lamina are small (50 nm in diameter) and may form small bundles called reticular fibers that stain with silver salts. The fibrils are covered with proteoglycan granules and are connected by ruthenium red-staining filaments that may consist of hyaluronic acid (see Chapter 2). Collagen fibers are larger than reticular fibers; they contain less proteoglycan and a greater variety of unit collagen fibrils (50–1500 nm in diameter). (From Hay *et al.*, 1978, referenced in Chapter 7.)

associated collagen fibrils, or reticular lamina) under an epithelium. Only in the glomerulus and a few other locations is the basal lamina free of collagen fibrils and thus truly equivalent to the basement membrane visualized previously by light microscopists. Another terminology problem is that in the glomerulus, the basal laminae of the epithelium and endothelium are fused, obliterating the layer we called the lamina rara interna above; the latter term is given to the PG-rich, juxtaendothelial zone of the glomerular basement membrane (GBM). In spite of the historical precedence of that use (in the GBM), most morphologists find it helpful to refer to the inner layer (facing connective tissue) of simple basement laminae as the internal lamina rara (Fig. i-1).

The term *connective tissue* is often used to refer to the collagen fibrils and other ECM molecules that surround cells of the fibroblast family. Some authors, however, include the basal lamina, as this structure is composed of molecules similar to those of the ECM proper and "connects" epithelium and muscle to the ECM proper. By connective tissue "cells," most authors mean cells of the fibroblast family (osteoblasts, chondroblasts, fibroblasts), but muscle and epithelial cells also secrete ECM and, in this sense, are part of the cellular component of connective tissue. Unlike the connective tissue cells proper (the fibroblast family), however, muscle and epithelial cells are separated from collagen fibrils by basal laminae; why this should be is not immediately obvious, but it is likely that these groups of cells have distinct cell surfaces that differ in their interactions with ECM (Chapter 12). Collagen fibrils are 10 nm in diameter or wider (above 25 nm in diameter, they appear striated); the term *filament* is usually used in this book to refer to a fibrous structure less than 10 nm in diameter. Collagen fibrils are often organized into small fibers called reticular fibers that are argyrophilic as seen in the light microscope, and into larger collagen fibers; the diameters of the fibrils composing these fibers vary in a predictable way (Fig. i-1).

*Cell Biology of Extracellular Matrix* is, as we noted above, a book written by experts, all of whom have contributed measurably to our understanding of the ECM. They have agreed to present their material succinctly and in a manner understandable and relevant to the cell biologist who has not been working in the field. The chapters are not "review" articles; for full coverage of historical and other details, I have asked the authors to refer the reader to the numerous, more technical reviews that appear in the journals and books of the trade and are readily accessible to the interested reader desiring more depth. This book is written for the cell biologist, then, but it does present the "state of the art" in ample detail to serve as a ready reference for all who wish to think and talk intelligibly about the ECM. Many of the techniques used to explore the ECM are presented in considerable detail, in footnotes, in figure legends, or in the text. The book is also fairly lavishly illustrated; I am a visual person myself and I have often added electron micrographs to the more biochemically oriented chapters. There is much in this book about disease, as well as health. This is, in short, a book that we believe will convince even the most doubting that to understand the cell is to understand the extracellular matrix.