Developmental Biology

A COMPREHENSIVE SYNTHESIS

Volume 5

The Molecular Biology of Cell Determination and Cell Differentiation

Edited by
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Cover illustration: Drosophila embryo that is homozygous for a strong mutation at the *Krüppel* locus. The embryo has been stained with an antibody to the *fushi tarazu* (*ftz*) segmentation protein. The *Krüppel* mutation causes an altered pattern of *ftz* stripes. (Micrograph courtesy of Dr. Sean Carroll, University of Wisconsin, Madison.)

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Grandma Kröte is enjoying the balmy summer night at the shores of her beloved pond. Unconcerned about science and scientists, she stubbornly keeps her secrets to herself. (Print by Dr. Johannes F. Holtfreter.)

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Preface

This series was established to create comprehensive treatises on specific topics in developmental biology. Such volumes serve a useful role in developmental biology, which is a very diverse field that receives contributions from a wide variety of disciplines. This series is a meeting ground for the various practitioners of this science, facilitating an integration of heterogeneous information on specific topics.

Each volume is comprised of chapters selected to provide the conceptual basis for a comprehensive understanding of its topic as well as an analysis of the key experiments upon which that understanding is based. The specialist in any aspect of developmental biology should understand the experimental background of the specialty and be able to place that body of information in context, in order to ascertain where additional research would be fruitful. The creative process then generates new experiments. This series is intended to be a vital link in that ongoing process of learning and discovery.

The application of the techniques and principles of molecular biology to problems of development has resulted in the most intense period of investigation and discovery in the history of developmental biology. Today, the major thrust of molecular biology is to understand the controls over differential gene expression. The identification of *cis*-acting regulatory sequences and demonstration of their potentiation by *trans*-acting factors have provided us with the framework for understanding how selective transcription of individual genes is mediated. Much remains to be learned, however, about the molecular mechanisms involved in activation and maintenance of transcriptional activity.

Developmental biologists have a broader mission, which is to explain how the complex program of differential gene expression is established and utilized during embryonic development. We recognize two general mechanisms for establishment of cellular domains having unique patterns of gene expression: segregation of cytoplasmic determinants into discrete mitotic derivatives and exposure of cells to extrinsic signals (i.e., embryonic induction). The roles of cytoplasmic determinants are discussed in Chapters 1 and 2. Embryonic induction has been studied most intensively in amphibians, beginning with Spemann's pioneering work. Johannes Holtfreter, Spemann's student, presents a personal perspective on Spemann's organizer in Chapter 4. Contemporary research on embryonic induction is discussed in Chapter 3.

Study of the expression of specific genes during development promotes the understanding of differential gene expression in a broader context, and the developmental systems highlighted in this book have been chosen with that aim in mind. Actin genes, discussed in Chapter 6, are expressed in lineage-specific patterns during sea urchin development. The coincidence of expression of specific actin genes and the establishment of lineages provide the opportunity to study cell determination at the molecular level with a precision never before possible. *Xenopus* ribosomal protein synthesis, discussed in Chapter 8, is subject to both transcriptional and posttranscriptional regulation during early development. Since selective transcription and delayed utilization of messenger RNA are both important strategies in animal development, this system provides an opportunity to learn how the embryo exploits these two regulatory modes to control protein synthesis.

Regulation of protein synthesis by steroids provides a means for ensuring that physiologically significant proteins will be rapidly produced by specific cells when needed by the organism. This strategy is particularly important for regulation of the reproductive cycle. This mode of regulation also provides the investigator with an important experimental advantage: the ability to modulate gene expression either *in vivo* or *in vitro* by the administration of exogenous hormone. The vitellogenin genes of *Xenopus*, discussed in Chapter 9, provide one of the best-studied examples of genes under steroid regulation.

There are two general strategies for studying gene control of development. One is to begin with the phenomenon and use molecular techniques to isolate the genes that are involved in its control. The cloned genes can then be experimentally modified and the effects of these modifications assessed. The other approach is to begin with a mutant gene that influences a developmental event. By comparison of the wild-type and mutant genes, the genetic basis for the defect and the regulation of the normal process can be determined. The latter approach has been particularly well exploited by investigators of *Drosophila* development. Genes affecting pattern formation of this insect have yielded a massive amount of information, which is masterfully discussed by Scott in Chapter 5. One of the best-studied gene loci in eukaryotes is the *rosy* locus of *Drosophila*, which encodes xanthine dehydrogenase (XDH). Mutant genes that have tissue-specific effects on XDH synthesis have been cloned, and the nucleotide sequences that exert this effect have been pinpointed. This process is discussed in Chapter 10. Further exploitation of this system should yield valuable information regarding the molecular mechanisms involved in tissue-specific gene expression.

In *Xenopus* development, the zygote is initially transcriptionally inert. Transcription is first detected at the mid-blastula stage. As discussed in Chapter 7, the rate of cell division influences the onset of transcription during *Xenopus* development. The relationship between mitosis and cell differentiation has fascinated developmental biologists for many years and is now under intense investigation. We shall soon learn a great deal about this relationship as it pertains to both normal development and malignant transformation.

One of the most surprising recent findings of molecular biology is that individual gene sequences may contain information that can be utilized to produce different proteins in different cells. The mammalian neuroendocrine system provides a remarkable example of differential processing of messengers to produce tissue-specific products, as is discussed in Chapter 11.

An important characteristic of cell determination is the stability of the determined state. Once determined to differentiate, cells normally retain their specificity, which they can transmit to their progeny at cell division. Any hypothesis of differential gene regulation must account for this fact. One means of achieving stability might involve heritable modifications to the nucleotides themselves. A correlation between hypomethylation of cytosine residues and transcriptional activity exists in higher animals and appears to play a role in determination and stability of differential transcription, as is discussed in Chapter 12.

Plant molecular biology has not yet yielded as much information as animal molecular biology, but it is emerging as an extremely important and active area of investigation, particularly due to the potential economic and social impact of the manipulation of plant gene expression. Mankind relies on plant proteins for food, clothing, and shelter. The ability to manipulate production of plant proteins has the potential to revolutionize the economies of both the developed and underdeveloped nations of the world. Part III of this book presents three of the finest systems for the study of differential gene expression in plants: Chapter 13 describes *Arabidopsis*, which has emerged as an outstanding organism for the conduct of genetic and molecular biological research; Chapter 14 discusses gene expression during seed development in flowering plants; and Chapter 15 deals with the synergistic relationship between plants and bacteria, which is necessary for nitrogen fixation. Not only is nitrogen fixation vital for nutrient enrichment, but it also provides an excellent experimental system for studying induced gene expression.

The publication of a book during a period of rapid progress, in which scientific journals report spectacular discoveries with incredible frequency, is risky. However, the authors have not attempted to present definitive answers to the problems of cell determination and differentiation. Rather, they bring us to the present level of understanding and lead us to the future, which is indeed going to be very exciting. I think they have succeeded admirably. I am proud of their achievements and I am confident that this book will serve both to inform its readers and to stimulate investigators to supplement our rapidly growing body of knowledge.

Leon Browder

Calgary, Alberta

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Specification of Regional Patterns of Cell Fate in Animal Development