

Embryonic Stem Cells

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Edited by

Kursad Turksen

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Preface

It is fair to say that embryonic stem (ES) cells have taken their place beside the human genome project as one of the most discussed biomedical issues of the day. It also seems certain that as this millennium unfolds we will see an increase in scientific and ethical debate about their potential utility in society.

On the scientific front, it is clear that work on ES cells has already generated new possibilities and stimulated development of new strategies for increasing our understanding of cell lineages and differentiation. It is not naïve to think that, within a decade or so, our overall understanding of stem cell biology will be as revolutionized as it was when the pioneering hemopoietic stem cell studies of Till and McCulloch in Toronto captured our imaginations in 1961. With it will come better methods for ES and lineage-specific stem cell identification, maintenance, and controlled fate selection. Clearly, ES cell models are already providing opportunities for the establishment of limitless sources of specific cell populations. In recognition of the growing excitement and potential of ES cells as models for both the advancement of basic science and future clinical applications, I felt it timely to edit this collection of protocols (*Embryonic Stem Cells*) in which forefront investigators would provide detailed methods for use of ES cells to study various lineages and tissue types.

We are pleased to provide *Embryonic Stem Cells: Methods and Protocols*, a broad-scaled work of 35 chapters containing step-by-step protocols suitable for use by both experienced investigators and novices in various ES cell technologies. In the first section of the volume, there are chapters with detailed protocols for ES cell isolation, maintenance, modulation of gene expression, and studies of ES cell cycle and apoptosis. *Embryonic Stem Cells* also includes chapters with protocols for the use of ES cells to generate diverse cell and tissue types, including blood, endothelium, adipocytes, skeletal muscle, cardiac muscle, neurons, osteoclasts, melanocytes, keratinocytes, and hair follicle cells. The second part of the volume contains a series of cutting edge techniques that have already been shown to have, or will soon have, tremendous utility with ES cells and their differentiated progeny. These chapters include the use of cDNA arrays in gene expression analysis, phage display antibody libraries to generate antibodies against very rare antigens, and phage display libraries to identify and characterize protein and protein interactions, to name a few. Collectively, these protocols should prove a useful resource not only to those who are using or wish to use ES cells to study fate choices and specific lineages, but also to those interested in cell and developmental biology more generally. We hope that this book will also serve as a catalyst spurring others to use ES cells for lineages not yet being widely studied with this model and to develop new methodologies that would contribute to both the fundamental understanding of stem cells and their potential utility.

Embryonic Stem Cells would not have materialized at all had the contributors not recognized the special value of disseminating their protocols and hard-won expertise. I am extremely grateful to them for their commitment, dedication, and promptness with submissions! I am also grateful to Dr. John Walker for having faith in and supporting me throughout this project. I wish also to acknowledge the great support provided by many at Humana Press, specifically Elyse O'Grady, Craig Adams, Diana Mezzina, and Tom Lanigan. A special thank you goes to my dedicated coworker, Tammy-Claire Troy, who, with her infectious optimism and tireless commitment, became a crucial factor in the editing and completion of the volume.

I am grateful to N. Urfe, P. Kael, and M. Chambers for their unintentional “awesome” contributions.

Finally, I hope that the volume will achieve the intent that I had originally imagined: that it will prove a volume with something for both experts and novices alike, that it will serve as a launching point for further developments in stem cells, and that we will all-too-soon wish to expand and update it with other emerging concepts, insights and methods!

Kursad Turksen

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- Plate 11 Fig. 1. (A, B) Transduction of mammalian cells by ligand-targeted phage.
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- Plate 12 Fig. 1. (A, B) Diagram illustrating the strategy for the selection of specific intracellular antibodies.
(See full caption and discussion on p. 435, Chapter 32.)
- Plate 13 Fig. 2. Diagram showing the restriction maps and polylinker sequences of the yeast expression vectors, (A) pBTM116 and (B) pVP16.
(See full caption and discussion on p. 437, Chapter 32.)
- Plate 14 Fig. 1. (A, B) Two alternative strategies to achieve complementation.
(See full caption and discussion on p. 448, Chapter 33.)
- Plate 15 Fig. 2. (A-H) Applications of the DHFR PCA to detecting the localization of protein complexes and quantitating protein interactions.
(See full caption and discussion on p. 451, Chapter 33.)
- Plate 16 Fig. 3. (A-C) β -Lactamase PCA using the fluorescent substrate CCF2/AM.
(See full caption and discussion on p. 455, Chapter 33.)