METHODS IN MOLECULAR BIOLOGY

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Mammary Stem Cells

Methods and Protocols

Edited by

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Preface

The concept of stem cells was first postulated more than a century ago, and over the last few years it has become a focus of interest and discussion even in nonscientific circles. The fact that this book is dedicated to Mammary Stem Cells reflects the attention that these cells have recently received and, as a consequence, many of the variety of approaches that researchers are using to investigate their properties and function are discussed here. Studying stem cells from the healthy mammary gland may not appear as relevant as the intriguing work presently being carried out with adult stem cells from other tissues, such as the heart or the nervous system, which epitomizes the great hope and potential for regenerative medicine in the future. However, understanding the role that stem cells play during development and in the physiology of the mammary gland is not only interesting in its own right but also essential to understand the molecular mechanisms underlying the alterations that lead to breast cancer, particularly when considering the hypothesis that the molecular basis of tumorigenesis is a stem/progenitor cell gone awry. Thus, the identification of normal and breast cancer stem cells more than a decade ago has offered a new vision of this heterogeneous disease and new hopes for its prognosis and treatment.

The collection of chapters in this book aims to guide the researcher interested in mammary stem cells, to isolate them and characterize them, at various different levels. Max Wicha, one of the protagonists in the field, opens the book with an extensive introduction to the subject that takes us from the nineteenth century to the current state of the art and a look into the future implications of targeting these cells in the clinic. Mammary cell lineage specification occurs during embryogenesis and thus Chapter 2 presents a protocol that allows the study of progenitor cells during embryonic mammary development ex vivo. The following two chapters provide guidance and experimental details that should help a researcher to start isolating stem cells using fluorescent-activated cell sorting. Once the stem/progenitor cells have been identified and isolated, there is a wide variety of methods to study them. Omics techniques are quickly becoming very informative tools, and there are three chapters that discuss DNA and mRNA sequencing, as well as proteomics techniques to assist in profiling cells in a more efficient manner. Chapter 8 focuses on lentiviral cell transduction, which provides an invaluable method to stably modify gene expression and facilitate functional studies. Transplantation studies have been considered the gold standard to demonstrate normal and cancer stem cell potential, and there are two chapters that provide detailed protocols to investigate mammary gland development and tumorigenesis by taking advantage of the mammary fat pads of mice. A more recent addition to the experimental alternatives to study stem cells is in vivo lineage tracing, which allows the researcher to track cell fate in situ, and this is discussed in detail in Chapter 11. Cancer stem cells not only function as tumor initiating cells but also can be implicated in resistance to therapy and disease dissemination. The finding that these cells display properties that allow them to establish themselves in a different environment is investigated in two chapters, which aim to follow stem cells from their initial niche to the new microenvironment at their metastasis site, to study their metastatic re-initiation capabilities and to establish an experimental system that allows monitoring of the survival, dormancy, and proliferation of

disseminated cancer cells. Finally, the last chapters are striking examples of how scientists are reaching out into other areas, from biology to physics and mathematics and back to biomedicine.

I hope that this book, by including some of the most basic techniques and exciting new developments, will help members of the scientific community new to the field to explore the behavior of stem cells and to learn how to tackle them. This knowledge should guide the design of new and complimentary strategies to be applied in the clinic with the final aim of fighting breast cancer.

Derio, Spain ***Maria del Mar Vivanco***

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