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469. Wnt Signaling, Volume 2: Pathway Models, edited by Elizabeth Vincan, 2008
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SARS- and Other Coronaviruses

Laboratory Protocols

Edited by

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Preface

The year 2003 was the year when the name “coronavirus” went around the world, somewhat further than the virus that sparked panic: severe acute respiratory syndrome coronavirus (SARS-CoV). It was spread rapidly by international, indeed transcontinental, travelers from its epicenter in China. The high mortality rate, around 10% among clinical cases, and the particularly high price paid by health care workers, spread fear globally. Public health facilities were stretched to the limit, and the effect on local economies was immense. Never before had a coronavirus made such an impact on the lives of the human population.

SARS enhanced interest in coronaviruses generally, including the hunt not only for SARS-CoV-like viruses in wild animals, but also for unrelated coronaviruses in wild and semi-domesticated animals. Prior to SARS we knew of only a dozen or so coronavirus species but it was always likely that there were many more. Coronavirologists knew, from decades of experience, that coronaviruses could be devastating, causing mortality, especially among young, and high economic loss among domestic animals. Moreover, they knew that a given coronavirus species was not limited to replication in one host species. Thus the potential existed for the spread of novel coronaviruses from wild animals to domestic animals and to man. It is with this potent threat in mind that I have included no fewer than seven chapters dealing with the detection and discovery of coronaviruses by nucleic acid approaches, with antibody-based approaches being described in three other chapters.

Although one can detect a virus these days without having to grow it in the laboratory, there is always a need to do this at some point, whether it be for the development of diagnostics and vaccines or for the study of pathogenesis, pathogenicity, variation, and other virological properties. Therefore, I have included several chapters on virus isolation and propagation.

For some purposes, e.g., structural studies (Section 3) and vaccine development, it is necessary to rigorously purify a virus after propagating it, an art that fewer virologists practice these days. Hence, two chapters contain detailed procedures for this. For other structural, and functional, studies and for raising antibodies for subsequent analytical use, it is sufficient, and, in the case of nonstructural proteins, essential, to express viral proteins individually. Several chapters address this task, including one that deals with crystallization of non-structural proteins.
Having antibodies specific to each viral protein is immensely useful for structure-function studies, but getting good antibodies is often problematic. Hence, I have included several chapters with protocols for raising antibodies against the viral proteins, including peptides as well as proteins expressed in vitro, and as part of whole virions.

The penultimate section of this book comprises five approaches to the manipulation of coronavirus genomes. These technical achievements have revolutionized the study of coronavirus replication and the development of vaccines against coronaviruses. SARS demonstrated that these approaches, developed with long-known coronaviruses, could be readily adapted to the molecular cloning, and subsequent manipulation, of a new coronavirus. These protocols are here to assist those who will rise to the challenge of new coronaviruses, and to provide hard-won practical advice for those working with current coronaviruses.

Finally, there are two chapters that describe how to investigate aspects of the cell surface receptors for coronaviruses. One of these deals with the sugar moieties that frequently play a role in virus attachment and can affect pathogenicity and tropism, but which tend to be overlooked once a protein has been identified as being a receptor. The other chapter describes how recombinant vesicular stomatitis virus can be used to study coronavirus receptors and tropism, safely, and offers an approach to vaccine development.

I would like to thank all the authors in this book for their devotion to the task. I am full of admiration for what they have achieved in their research and their willingness to describe their protocols in minute detail, including the sort of practical advice that is never included in primary publications.

Although the protocols focus on coronaviruses, it seems to me that all the chapters have something to offer every virologist. Indeed, I believe that, among us, we have produced a protocol book for virologists in general, as well as for those, present and future, who, through choice or circumstance, work with coronaviruses.

Dave Cavanagh
Compton
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A Guide to the Sections of this Book

Section 1: Detection and Discovery of Coronaviruses

See the following chapters, especially, for the use or detection of antibodies to detect coronaviruses:

Hasoksuz et al. (Section 1) for descriptions of ELISAs for detection of antibody and of antigen.

Lambert et al. (Section 2), for detection of coronaviruses in cell culture, using indirect immunoperoxidase staining.

Ohnishi (Section 5), for virus detection by immunofluorescence, immunoblotting, and antigen capture ELISAs.

Section 2: Isolation, Growth, Titration and Purification of Coronaviruses

See the following chapters also:

Hasoksuz et al. (Section 1) for virus growth and plaque titration.

Neuman and Buchmeier (Section 3) for virus purification.

De Haan et al. (Section 6) for virus propagation and plaque titration.

Donaldson et al. (Section 6) for virus propagation and plaque titration.

Schwegmann-Wessels and Herrler (Section 7) for virus propagation and purification.

Section 4: Expression of Coronavirus Proteins, and Crystallization

See also the following chapters:

Zevenhoven-Dobbe et al. (Section 5) for expression of coronavirus proteins in Escherichia coli.

Pendleton and Machamer (Section 5) for expression of coronavirus proteins in Escherichia coli.