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Immunophenotyping

Methods and Protocols

Edited by

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Humana Press

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ISSN 1064-3745 ISSN 1940-6029 (electronic)
Methods in Molecular Biology
ISBN 978-1-4939-9649-0 ISBN 978-1-4939-9650-6 (eBook)
https://doi.org/10.1007/978-1-4939-9650-6

This Humana imprint is published by the registered company Springer Science+Business Media, LLC, part of Springer Nature.
The registered company address is: 233 Spring Street, New York, NY 10013, U.S.A.

Preface

Immunophenotyping has become more complex in recent years due to the increasing availability of new fluorochromes with novel spectral characteristics and the development of instrumentation with expanded capabilities. Propelled by technological advances in cytometry as well as the development of commercially available monoclonal antibodies, immunophenotyping of a multitude of cell types using flow cytometry has become commonplace in virtually all academic research centers, pharmaceutical companies, and clinical laboratories. Thirty-five to forty years ago, immunophenotyping was limited to one or two colors while today it is possible to construct panels consisting of several dozen fluorescent-conjugated markers. Similarly, analysis of flow cytometric immunophenotyping has developed beyond real time, direct observation of electronic signals represented by one- or two-parameter histograms viewed at the cytometer. Today, data acquired in list mode fcs format files allows for remote, retrospective, highly complex, and sometimes automated, data analysis performed independent of the cytometer and shared beyond the laboratory. Additionally, newer cytometric technologies such as imaging flow cytometry and mass cytometry permit immunophenotyping in conjunction with morphologic studies or with an expanded number of parameters, respectively. The following chapters will present a number of these topics as well as newer developments in the field of immunophenotyping.

Today, it is virtually impossible to present a complete and comprehensive compendium of all immunophenotyping methods and applications in one volume. Therefore, this text will present a representative collection of immunophenotypic methods and applications with examples of newer technologies and reagents used in the research and clinical environments. Basic methods in immunophenotyping such as construction of high-dimensional fluorescence and mass cytometry panels; fluorescence barcoding; and use of dried or lyophilized reagents will be presented first, followed by immunophenotyping examples of specific cell types often studied in research laboratories. The final chapters present immunophenotyping topics useful in the clinical laboratory, including a chapter on the critical role of quality control and immunophenotyping in the clinical environment.

I would like to thank each of the authors for volunteering to take the time to share their expertise for this volume. Without their hard work and dedication, it would not have been possible to share these methods with you.

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