

polymorphism of the involved receptors and the role of natural killer cells, there is now better realization on the important and clinically meaningful variations that underlie the innate responses. Of greater interest is the observed inter-individual variation in immune response leading to differential disease susceptibility and allograft success across ethnic groups and populations. The challenge is to develop robust and easy to do assay systems for quantitative analysis of the immune response with desired degree of efficiency and accuracy.

This handbook is a compendium of experimental approaches for investigating various components of the human innate immunity at molecular and subcellular level, presented in an easy to follow format. Edited by two practitioners of immunology with years of hands-on experience, the book presents a practical refresher course on the recently developed high throughput and advanced technologies to investigate functional characteristics of the innate immune pathways. The content has been contributed by experienced scientists from important institutions with focus on rationale, practical tips, applications and advantages of the various high throughput techniques. There is clear presentation of standardized protocols used to investigate various aspects of the innate immune system, particularly the Toll-like receptor (TLRs) expression and function, NK (natural killer) cells, neutrophils, mononuclear cells, phagocytes and others.

The book is compiled into 12 chapters in all. The first five chapters include characterization of innate immune system using flow cytometry, high density single cell mass cytometry, (CyTOF), high throughput secretomic analysis of single cell, decellularization techniques, imaging of rare cell populations and interaction of innate immune system by ImageStream methodology. The chapters 6-9 are focused on molecular characterization using various molecular methods and their protocols like transcriptomic profiling, miRNA and RNAi based analysis to elucidate innate immune signal transduction. Finally, the chapters 10-12 are comprised of analytical approaches including statistical analysis, system approaches in autoimmune diseases and genetic mapping of immune system function. The latter has emerged as an important tool to define the genetic and phenotypic complexity of the immune system in relation to understanding disease pathogenesis.

This book gives an overview of the important advancements and evolution of immunological

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With new information on the innate immune pathways for cellular recognition, genetic

techniques used in investigating the innate immune system. The chapters are informative with relevant literature particularly on the use of microfluidic approaches, single cell mass cytometry and bioinformatics. The high dimensional single cell mass cytometry is a recently developed technique for dissecting complex cellular systems with advancement in traditional flow cytometry. It is a next generation flow cytometry that works with an instrument, CyTOF (cytometry by Time-Of-Flight) mass spectrometer with advanced unsupervised tools, SPADE and viSNE. This technique works on the basis of the use of antibodies conjugated to metal isotopes rather than fluorophores that ultimately help in detection of more than 38 parameters and help in characterization of a single cell within a complex heterogenous population of cells with overlapping cell markers in various differential stages. The characterization of mononuclear phagocyte system, particularly their polarized subsets, tumour associated macrophages (TAM) and myeloid derived suppressor cells (MDSCs), on the basis of their phenotypes, functions and lineage identity by the use of high dimensional single cell mass cytometry, is well explained.

Additionally, another chapter explains the secretomic analysis of single cells to assess the cellular

heterogeneity. This chapter describes the use of a new microfluidic assay for high throughput, multiplexed measurements in single living cells. This technique is designed to handle very small volumes of the sample. While the conventional assays measure responses in cell populations, obscure subsets of innate immune cells that may have distinct secretory functions are not that easy. Additionally, other single cell based assays like ELISpot and intracellular cytokine staining (ICS) by flowcytometry also have limitations similar to these assays, for instance, cells cannot be recovered for further analysis after the assay is complete.

Overall this book is a useful addition to the shelf for students of immunology, particularly young researchers who are keen to understand the basics and applications of the most advanced techniques in investigating the innate immune system. This book is recommended to all libraries and immunology research groups.

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