
Preface

Proteins are one of the most important classes of molecules in living cells and tissues. Proteins and their complexes are involved in virtually all cellular processes including catalysis of biochemical reactions, transport of molecules across membranes, cell growth and division, cell adhesion and migration, and providing structural support. Characterizing the quantity, association, and activity of proteins is therefore critical for understanding the molecular mechanisms of cellular processes involved in cell differentiation and fate, cell signaling, disease progression, and for discovery and development of novel therapeutics, vaccines, and diagnostics. Measuring DNA and RNA can provide qualitative information on gene products (proteins) but cannot provide information on protein concentration, activity, location, posttranslational modifications, or interactions with other proteins. Therefore, we need tools and assays to directly measure proteins, their interactions and activities, and modifications. Numerous analytical methods have been developed to analyze proteins such as gel electrophoresis, immunoassays, enzyme assays, chromatography, and mass spectrometry. At the conventional scale however, these methods require a large number of cells for analysis, resulting in a population-averaged measurement. Cells are heterogeneous in nature and even genetically identical cells exhibit heterogeneous behavior. This heterogeneity may have important biological consequences for both the individual cells and the population. Population-averaged data, which assumes that all the cells are identical, can be misleading and hence more desirable data in many instances is data from analyses of single cells. One well-known example is the response of bacteria to antibiotics where at certain doses many cells die but some survive and develop resistance. Similarly, one of the unanswered questions in cancer therapy has been why essentially identical cells respond differently to a drug. Single-cell level measurement of proteins (and other molecules) has provided valuable insight into mechanisms that dictate heterogeneity in cellular response to drugs and other internal and external stimuli. Usefulness of single-cell measurements is also obvious for stem cell research as decisions in individual cells determine their fate.

Despite the need, tools that allow quantitative measurements of proteins in single cells have not been easily available. The biggest challenge to measuring proteins in single cells is the exceedingly low amount present in a cell. The complexity and large concentration range (fM to high nM) of the proteome add additional challenges. Significant efforts are currently being made to overcome these challenges and achieve selective and sensitive analysis of the proteome in individual cells. This volume highlights recent developments in flow cytometry, affinity assays, imaging, mass spectrometry, microfluidics, and other technologies that have enabled analysis of the proteins at the single-cell level. We also include chapters covering a suite of biochemical and biophysical methods capable of making the entire gamut of proteomic measurements including analysis of protein abundance or expression, protein interaction networks, posttranslational modifications, translocation, and enzymatic activity. The book should be useful to researchers and students in biological and biomedical sciences who have an interest in proteomic measurements in cells.

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