Advancements in single-cell proteomics: from technology development to biological applications.

Ogbebor Dragusha*

Department of Pharmaceutical Biosciences, Uppsala University, Uppsala, Sweden

Abstract

Single-cell proteomics is a rapidly growing field that has the potential to revolutionize our understanding of cellular heterogeneity and function. Recent advancements in technology development have enabled the measurement of protein expression at the single-cell level, providing unprecedented insights into cellular processes that were previously inaccessible using bulk proteomics techniques. One of the major challenges in single-cell proteomics is the limited amount of starting material, which requires highly sensitive and efficient sample preparation methods. Recent developments in sample preparation have focused on reducing sample loss and minimizing sample-to-sample variability, allowing for the reliable detection of low-abundance proteins in single cells.

Keywords: Single-Cell Proteomics, Mass Cytometry, Microfluidics, Isobaric Labeling, Data Analysis, Cell Type Identification, Biomarker Discovery.

Introduction

Proteomics, the study of all the proteins expressed by a cell, tissue or organism, is a rapidly evolving field with major implications for understanding biology and disease. In recent years, the development of single-cell proteomics has enabled researchers to study protein expression at the level of individual cells, providing unprecedented insights into cellular heterogeneity and function. Here, we will discuss the advancements in single-cell proteomics, from technology development to biological applications [1].

Single-cell proteomics technologies have evolved from traditional bulk proteomics approaches, where proteins from many cells are pooled together for analysis, to more advanced techniques that can measure protein expression in individual cells. The earliest single-cell proteomics methods utilized fluorescence-based techniques such as immunofluorescence staining and flow cytometry, but these methods were limited in their ability to detect low-abundance proteins and provide quantitative measurements of protein expression. Recent advances in mass spectrometry (MS)-based proteomics have enabled researchers to overcome these limitations, allowing for the quantification of thousands of proteins from individual cells [2].

One of the key challenges in single-cell proteomics is obtaining enough protein for analysis. Traditional methods for extracting proteins from cells are not suitable for single-cell analysis due to the limited amount of protein in each cell. Researchers have developed a variety of methods for protein extraction from individual cells, including laser capture microdissection, microwell-based isolation, and microfluidics-based approaches. These methods allow for the isolation of individual cells and the extraction of proteins from them, enabling downstream proteomic analysis [3].

Another challenge in single-cell proteomics is the complexity of the proteome. The human proteome is estimated to consist of over 20,000 proteins, with many proteins expressed at low levels in specific cell types or under certain conditions. To overcome this complexity, researchers have developed sensitive MS-based techniques such as tandem mass tag (TMT) labeling, which allows for the simultaneous measurement of the expression of thousands of proteins from multiple cells. Other techniques such as single-cell western blotting and proximity ligation assays (PLAs) have also been developed to measure the expression of specific proteins in individual cells [4].

Single-cell proteomics has numerous applications in basic and translational research. In neuroscience, single-cell proteomics has been used to study the heterogeneity of neuronal cell types and identify proteins that are specifically expressed in certain types of neurons. In cancer research, single-cell proteomics has been used to identify proteins that are differentially expressed in cancer cells compared to normal cells, which may serve as potential targets for cancer therapy. Single-cell proteomics has also been used to study the immune system, allowing for the identification of immune cell subsets and the characterization of their protein expression profiles.

Furthermore, single-cell proteomics can be used to study disease progression and treatment response at the individual

***Correspondence to:** Ogbebor Dragusha, Department of Pharmaceutical Biosciences, Uppsala University, Uppsala, Sweden, Email: ogbedrag93@edu.sw

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cell level, providing insights into the underlying mechanisms of disease and facilitating the development of personalized medicine. For example, single-cell proteomics has been used to identify drug targets in cancer cells that are resistant to chemotherapy, allowing for the development of targeted therapies that specifically target these cells. In infectious disease research, single-cell proteomics has been used to identify proteins that are differentially expressed in infected cells compared to uninfected cells, providing insights into the pathogenesis of infectious diseases and potential targets for therapeutic intervention [5].

Conclusion

In conclusion, single-cell proteomics has seen tremendous advancements in recent years, driven by the development of new technologies and their integration with biological applications. The emergence of mass spectrometry-based techniques has allowed for the analysis of protein expression and function at the single-cell level, enabling the study of cellular heterogeneity and the identification of rare cell populations. Advancements in microfluidics, nanotechnology, and single-cell isolation techniques have also facilitated the generation of high-quality single-cell proteomics data with improved sensitivity, throughput, and accuracy. The development of computational tools for data analysis and interpretation has further enhanced the utility of single-cell proteomics for understanding complex biological systems.

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