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Single-cell Proteomics: Deciphering Protein Turnover in Mammalian Cells

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ABSTRACT

Single-cell technologies have revolutionized modern biology, with early advances driven primarily by high-throughput sequencing and imaging platforms. Among emerging approaches, single-cell proteomics has gained considerable attention because proteins represent the principal functional effectors of cellular physiology. Protein turnover, the dynamic equilibrium between protein synthesis and degradation, is fundamental to maintaining cellular homeostasis, regulating signalling pathways, and preserving cell viability. Over the past decade and particularly in the last five years, substantial technological progress has accelerated the development of single-cell proteomics, despite the intrinsic analytical challenges posed by the lack of protein amplification strategies comparable to those available for nucleic acids. Increasing evidence demonstrates that single-cell proteomics provides critical complementary information to single-cell transcriptomics, enabling a more comprehensive understanding of cellular function and heterogeneity. This review summarizes the current landscape of single-cell proteomics and key biological applications. It highlights recent advances that enable sensitive analysis of protein turnover dynamics in individual cells and complex populations, as well as major technical and computational challenges, including limited sample volumes, sensitivity, reproducibility, and normalization.

INTRODUCTION

Protein turnover is a fundamental biological process involving the continuous synthesis and degradation of proteins, thereby maintaining cellular proteostasis, regulating signalling pathways, and enabling adaptive responses to environmental and physiological changes (de Souza et al., 2025). Through the selective removal of damaged, misfolded, or redundant proteins and their replacement with newly synthesised molecules, protein turnover preserves cellular integrity and function (Ahmad and Budnik, 2023). Disruption of this tightly regulated balance contributes to the onset and progression of numerous pathological conditions, including cancer, neurodegenerative disorders, and metabolic diseases (Acosta et al., 2025). Protein turnover encompasses the dynamic balance between protein synthesis and degradation and represents a critical determinant of cellular homeostasis. Unlike static measurements of protein abundance, turnover analyses offer time-based insights into proteome dynamics, enabling the investigation of how cells respond to stress, environmental stimuli, disease progression and therapeutic interventions. In recent years, substantial advances in stable isotope labelling strategies, high-resolution MS, and quantitative proteomics have significantly improved the ability to measure protein turnover across diverse biological systems (Labib and Kelley, 2020). Several isotope-based metabolic labelling approaches have become central to turnover studies, including Stable Isotope Labelling by Amino Acids in Cell Culture (SILAC), dynamic SILAC, and metabolic incorporation of isotopes such as ^15N and deuterium oxide (D_2O). These methodologies enable the tracking of newly synthesised proteins through incorporation of isotopically labelled precur-

sors, thereby facilitating quantitative estimation of protein synthesis rates, degradation kinetics, and protein half-lives. Coupled with high-resolution MS and data-independent acquisition (DIA) workflows, these approaches have markedly enhanced analytical sensitivity, accuracy, and reproducibility, even in complex tissues and low-abundance proteomes. A major advancement in the field has been the emergence of single-cell proteomics for the investigation of protein turnover dynamics. Conventional bulk proteomic analyses provide only averaged measurements across heterogeneous cell populations, thereby obscuring cell-to-cell variability within tissues. However, recent developments in nanoproteomics, microfluidics, and ultra-sensitive MS platforms now permit proteomic measurements at single-cell resolution. Technologies such as Single-Cell Proteomics by Mass Spectrometry (SCoPE-MS) and related nano-scale proteomic workflows enable quantitative analysis of protein turnover within individual cells (Werner et al., 2024). These approaches are particularly valuable for studying tumour heterogeneity, immune-cell dynamics, and rare cellular subpopulations that exhibit distinct proteomic and metabolic states.

In addition, multiplexed tandem mass tag (TMT)-based labelling strategies combined with carrier proteomes have substantially increased throughput and quantification accuracy in single-cell proteomics. Integration of microfluidic systems and spatially resolved proteomic technologies has further enabled longitudinal and spatial analyses of protein turnover using minimal sample inputs. Collectively, these innovations represent a transformative shift in dynamic proteomics, enabling high-resolution and time-resolved investigation of cellular protein dynamics across experimental models and clinical specimens (Sabatier et al., 2025). Protein turnover studies have generated critical biological insights into protein function and cellular regulation. Turnover kinetics distinguish short-lived regulatory proteins from long-lived structural proteins and reveal disease-associated alterations in protein stability, folding, and degradation pathways. Changes in protein turnover rates can indicate abnormal proteostasis, impaired protein quality-control mechanisms, or altered ubiquitin-proteasome and autophagy pathways, all of which contribute to disease pathogenesis, tumour progression, therapeutic resistance, and pathological protein aggregation (Marinko et al., 2019). From a clinical perspective, protein turnover analysis is increasingly recognised as a powerful approach for biomarker discovery, therapeutic monitoring, and precision medicine. Unlike conventional proteomic measurements that quantify static protein abundance, turnover-based analysis provides dynamic information regarding disease activity, treatment response, and patient-specific molecular phenotypes. Such approaches hold significant promise for identifying early molecular alterations and improving personalised therapeutic strategies. In cancer, dysregulated protein turnover promotes uncontrolled cellular proliferation, tumour progression, and therapeutic resistance by stabilising oncogenic proteins or accelerating the degradation of tumour suppressors. Similarly, in neurodegenerative diseases such as Alzheimer's and Parkinson's disease, the impairment of proteolytic pathways results in the accumulation of toxic aggregation-prone proteins, contributing to neuronal dysfunction and degeneration. Metabolic disorders, including diabetes mellitus and non-alcoholic fatty liver disease, are also associated with aberrant protein turnover pathways that alter enzyme stability and intracellular signalling networks (Bou Matar et al., 2025). Several challenges remain, including sample complexity, limitations in proteome coverage, data integration, standardisation of analytical workflows and computational interpretation of large-scale turnover datasets. Nevertheless, the convergence of isotope labelling methodologies, advanced MS technologies and single-cell proteomics is rapidly establishing protein turnover analysis as a central component of dynamic proteomics and translational biomedical research. (Labib and Kelley, 2020). This review therefore highlights the methodologies, biological applications and future perspectives of protein turnover research in both experimental and clinical settings.

SAMPLE PREPARATION FOR SINGLE-CELL PROTEIN TURNOVER

Sample preparation is one of the most critical steps in single-cell proteomics because a single mammalian cell typically contains only 100-500 pg of total protein, making protein losses during processing a major analytical challenge (Ahmad and Budnik, 2023). Unlike nucleic acids, proteins cannot be amplified; therefore, highly efficient, low-loss sample-preparation workflows are essential for obtaining reliable quantitative data. Recent advances in miniaturized processing platforms, microfluidics, nanodroplet technologies, and automated liquid handling have substantially improved protein recovery, reproducibility, and sensitivity in single-cell proteomic analyses (Labib and Kelley, 2020; Sabatier et al., 2025).

Single-cell isolation

The first step involves the isolation of individual cells from cultured cells, tissues, or biological fluids while preserving cellular integrity and minimizing contamination. Several isolation techniques are currently employed, including fluorescence-activated cell sorting (FACS), magnetic-activated cell sorting (MACS), laser capture microdissection (LCM), micromanipulation, microfluidic devices, and droplet-based platforms. The choice of isolation method depends on the biological sample, cell abundance, and downstream analytical requirements. Microfluidic systems are increasingly preferred because they reduce sample loss, improve throughput, and enable automated processing of individual cells (Liu et al., 2019; Ahmad and Budnik, 2023).

Cell lysis and protein extraction

Following isolation, individual cells are lysed using MS-compatible buffers containing mild detergents or surfactants that efficiently release intracellular proteins while minimizing protein degradation. Mechanical disruption, freeze-thaw cycles, thermal lysis, or chemical lysis are commonly employed depending on the sample type. Because of the extremely low protein content, sample preparation protocols are designed to minimize protein adsorption to reaction vessels and pipette surfaces. Miniaturized reaction volumes and nanoliter-scale processing significantly improve protein recovery and analytical sensitivity (Labib and Kelley, 2020).

Protein reduction, alkylation, and enzymatic digestion

Extracted proteins are reduced using reagents such as dithiothreitol (DTT) or tris (2-carboxyethyl) phosphine (TCEP) to break disulfide bonds, followed by alkylation with iodoacetamide (IAA) or chloroacetamide to prevent disulfide bond reformation. Proteins are subsequently digested into peptides using sequence-specific proteases, primarily trypsin, although Lys-C is frequently used either alone or in combination with trypsin to improve digestion efficiency. Complete and reproducible digestion is essential for accurate peptide identification and quantitative protein turnover analysis.

Peptide cleanup and labelling

Following digestion, peptides are purified to remove salts, detergents, and other contaminants that interfere with mass spectrometric analysis. Solid-phase extraction (SPE), stage tips, magnetic bead-based methods such as SP3 (Single-Pot Solid-Phase-enhanced Sample Preparation), and nanodroplet processing platforms are commonly employed for peptide cleanup. For multiplexed single-cell proteomics, peptides are chemically labelled using Tandem Mass Tags (TMT), enabling simultaneous analysis of multiple single cells together with carrier proteomes, thereby improving peptide identification and quantitative accuracy (Ghosh et al., 2025).

Miniaturized and automated sample preparation platforms

Recent technological developments have transformed single-cell sample preparation through miniaturized and automated workflows. Platforms such as nanoPOTS (Nanodroplet Processing in One Pot for Trace Samples), mPOP (minimal ProteOmic sample Preparation), autoPOTS, and integrated microfluidic systems substantially reduce sample loss by performing all preparation steps in nanoliter-scale reaction volumes. These technologies improve protein recovery, analytical reproducibility, throughput, and compatibility with ultra-sensitive LC–MS/MS platforms, thereby facilitating large-scale single-cell protein turnover studies (Sabatier et al., 2025; Olukotun, 2025).

Quality control considerations

Quality control during sample preparation is essential for obtaining reproducible single-cell proteomic data. Critical parameters include cell viability, contamination control, digestion efficiency, peptide recovery, batch effects, and technical reproducibility. The incorporation of internal standards, carrier proteomes, automated liquid handling, and standardized protocols significantly improves quantitative accuracy and reduces technical variability across experiments. Careful optimization of these parameters is particularly important for protein turnover studies, where accurate quantification of isotopically labelled peptides directly influences the estimation of protein synthesis rates, degradation constants, and protein half-lives.

METHODS OF SINGLE-CELL PROTEIN TURNOVER ANALYSIS

Stable isotope labelling is a fundamental approach for studying protein turnover, enabling precise quantification of protein synthesis and degradation rates in living systems. This technique involves the incorporation of non-radioactive heavy isotopes, such as ^13C, ^15N, and ^2H, into amino acids or metabolic precursors that are subsequently incorporated into newly synthesised proteins. The labelled proteins can then be

distinguished from pre-existing proteins using high-resolution mass spectrometry, enabling monitoring of changes in protein abundance over time. By measuring the rate of isotope incorporation during the labelling phase and its disappearance during the chase period, protein synthesis rates, degradation kinetics and protein half-lives can be accurately determined (Claydon et al., 2012). Several stable isotope-based methodologies have been developed to address different experimental requirements. Stable isotope labelling by amino acids in cell culture (SILAC) is widely used for cultured cells, whereas heavy water (^2H2O) labelling and ^15N labelling are commonly employed in animal studies. These approaches provide quantitative insights into proteome dynamics under physiological and pathological conditions and have greatly enhanced our understanding of cellular regulation, adaptation, and disease mechanisms (Lill, 2003). Combined with advances in mass spectrometry instrumentation and computational analysis, stable isotope labelling has become an indispensable tool for investigating protein turnover at both the individual protein and mixed protein levels.

Stable isotope labelling by amino acids in cell culture (SILAC)

Stable isotope labelling by amino acids in cell culture (SILAC) is one of the most widely used metabolic labelling approaches in quantitative proteomics. In SILAC, cells are cultured in media supplemented with amino acids labelled with stable heavy isotopes, most commonly ^13C- or ^15N-labelled lysine and arginine (Fig. 1; Bernhardt et al., 2025). During protein synthesis, these labelled amino acids are incorporated into nascent proteins, while pre-existing proteins retain light (unlabelled) amino acids. In pulse-chase SILAC experiments, cells are switched between labelled and unlabelled media, allowing temporal monitoring of protein synthesis and degradation. The relative abundance of heavy and light peptide species is quantified by mass spectrometry, enabling calculation of protein turnover rates and half-lives. SILAC offers high quantitative accuracy and minimal sample-processing variability because labelled and unlabelled proteins are combined prior to downstream processing. Consequently, SILAC has been extensively applied to investigate proteome remodelling during cellular differentiation, stress responses, drug treatment, and disease progression.

Dynamic SILAC is an extension of conventional SILAC designed for time-resolved analysis of proteome turnover. In this approach, cells are exposed to isotopically labelled amino acids for defined intervals, and samples are collected at multiple time points following label incorporation (Alevra et al., 2019). Temporal changes in heavy-to-light peptide ratios are then analysed to generate protein-specific turnover curves. This strategy enables estimation of kinetic parameters, including protein synthesis rates, degradation constants, and protein half-lives, at a proteome-wide scale. Integration of dynamic SILAC datasets with computational and mathematical modelling further facilitates the identification of post-translational regulatory mechanisms and pathway-specific proteome adaptations. Dynamic SILAC has proven particularly valuable for studying rapid proteomic responses associated with signalling events, cell-cycle progression, stress adaptation, and pathological conditions.

Although SILAC is highly effective in cultured cells, its application is limited in whole-organism studies. To overcome this limitation, metabolic labelling approaches using ^15N or deuterium oxide (D_2O) have been developed for in vivo analysis of protein turnover in animal models and humans (Bernhardt et al., 2025). In ^15N metabolic labelling, organisms are supplied with ^15N-enriched amino acids or nitrogen sources through diet or culture media, resulting in global incorporation of ^15N into newly synthesised proteins. This method enables long-term and tissue-specific turnover measurements and has been widely used in developmental biology, ageing research, and disease progression studies. D_2O labelling involves the administration of heavy water either orally or via injection. Deuterium from body water becomes incorporated into non-essential amino acids through intermediary metabolism and is subsequently integrated into newly synthesised proteins. Compared with amino acid-based labelling strategies, D_2O labelling is minimally invasive, cost-effective, and suitable for long-term studies in humans. It has therefore been extensively utilised in clinical investigations of muscle protein synthesis, metabolic disorders, cardiovascular diseases, and cancer biology (Lei et al., 2025). Collectively, ^{15}N and D_2O labelling methods provide powerful tools for investigating protein dynamics under physiological and pathological conditions in complex biological systems, thereby enhancing the translational relevance of protein turnover studies.

Mass spectrometry-based proteomics

Mass spectrometry (MS)-based proteomics constitutes the principal analytical platform for protein turnover studies. Coupled with stable isotope labelling strategies, MS enables highly sensitive identification and quantitative measurement of isotopically labelled peptides and proteins (Lill, 2003). Advances in MS

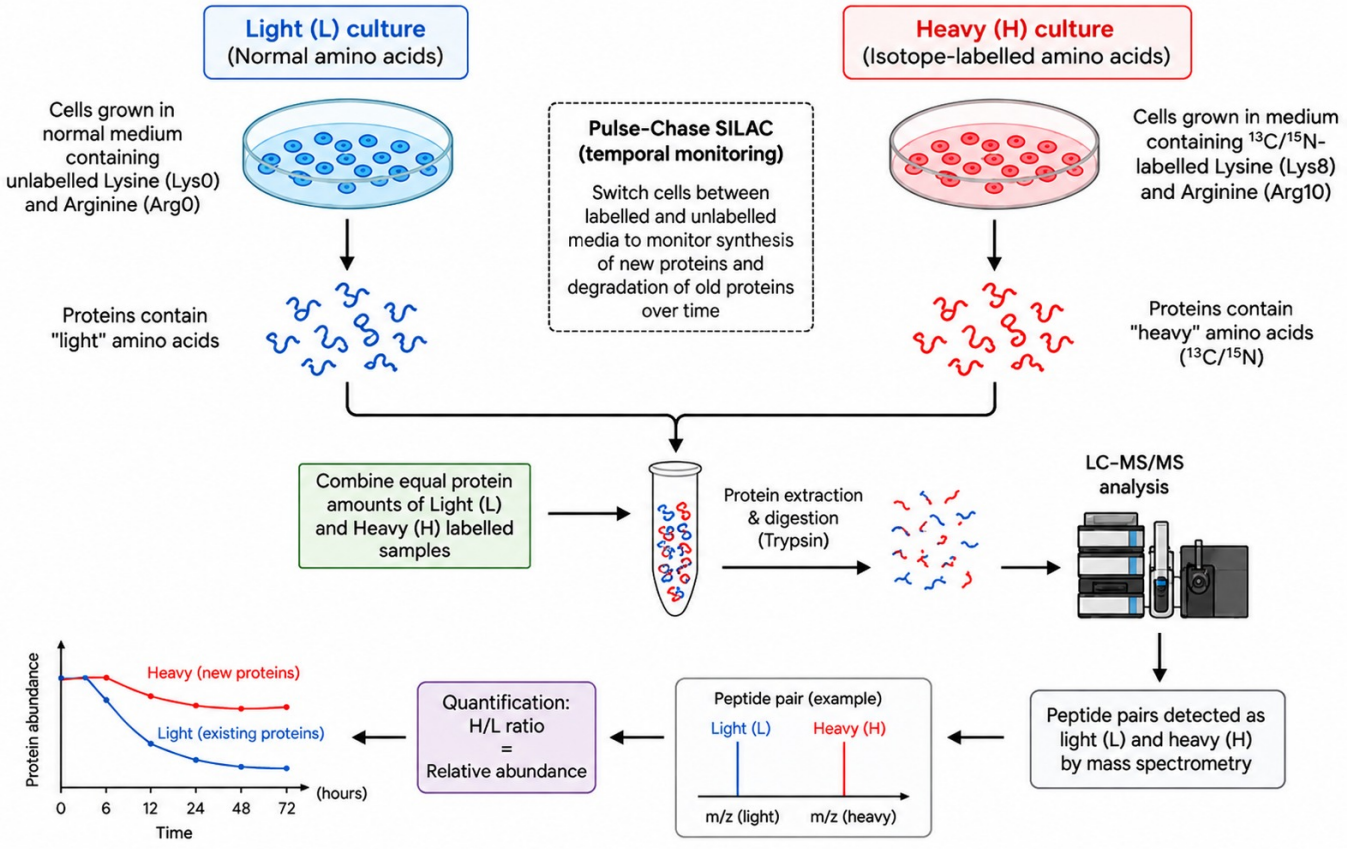

Figure 1: Workflow for stable isotope labelling of amino acids in cell culture (SILAC).

instrumentation and acquisition methodologies have substantially improved the accuracy, depth, and reproducibility of turnover analyses, facilitating large-scale characterisation of proteomic dynamics. High-resolution MS instruments, including orbitrap and time-of-flight (TOF) analysers, provide accurate mass determination and high resolving power for peptide analysis. These instruments can precisely detect mass differences generated by the incorporation of stable isotopes such as ^13C, ^15N, and ^2H into newly synthesised proteins (Liu et al., 2007). Detection of isotope-induced mass shifts enables discrimination between newly synthesised (labelled) and pre-existing (unlabelled) protein populations. Quantitative analysis of labelled-to-unlabelled peptide ratios permits accurate estimation of protein synthesis rates, degradation kinetics, and turnover half-lives across thousands of proteins simultaneously. High-resolution MS also facilitates detection of subtle alterations in protein turnover associated with cellular stress, ageing, developmental transitions, and disease states. Recent technological improvements in ion transmission efficiency, scan speed, and mass accuracy have further increased proteome coverage and analytical sensitivity, enabling turnover analysis of low-abundance proteins and limited biological samples. Data-independent acquisition (DIA) has emerged as a powerful MS acquisition strategy for quantitative proteomics and protein turnover analysis, ensuring comprehensive and reproducible peptide acquisition (Guarnaccia et al., 2025).

Single-cell proteomics by mass spectrometry (SCoPE-MS)

The study of protein turnover at the single-cell level represents a major breakthrough in proteomics, enabling the investigation of cellular heterogeneity that is often obscured in bulk population analyses (Potthoff et al., 2025). Recent advances in single-cell mass spectrometry, spatial proteomics, and integrated imaging technologies now permit highly sensitive quantification of protein synthesis and degradation within individual cells. These approaches are particularly valuable for analysing complex and heterogeneous biological systems, including tumours, immune microenvironments, and early embryonic development, where distinct cellular subpopulations exhibit unique metabolic and functional states. Single-cell proteomics by mass spectrometry (SCoPE-MS) is a pioneering approach that combines tandem mass tag (TMT) multiplexing with carrier proteomes to enable proteomic analysis of individual cells. Tandem mass tag (TMT) labelling is a quantitative proteomics strategy in which peptides from different biological samples are chemically la-

belled with isobaric tags that generate unique reporter ions during MS/MS fragmentation. TMT reagents enable simultaneous analysis of up to 16 samples in a single MS experiment, and the addition of a carrier proteome, consisting of peptides derived from bulk cell populations, enhances peptide identification and fragmentation efficiency by increasing ion signal intensity (Olukotun, 2025). In turnover studies, SCoPE-MS facilitates the discrimination between newly synthesised and pre-existing proteins at single-cell resolution. This capability enables the identification of turnover heterogeneity among cellular subpopulations, such as stem cells versus differentiated cells, therapy-sensitive versus resistant cancer cells, or activated versus quiescent immune cells. Consequently, SCoPE-MS provides important insights into dynamic cellular adaptation, lineage commitment, and disease progression.

Nanoproteomics

Nanoproteomics encompasses highly sensitive analytical workflows specifically designed for extremely limited sample quantities, often in the picogram-to-femtogram range. These approaches incorporate miniaturized sample preparation systems, including nano-liquid chromatography (nanoLC), microfluidic platforms, and ultrasensitive MS technologies. Coupling nano-proteomics with metabolic labeling strategies, such as dynamic SILAC, further enhances the ability to quantify protein turnover in minute cellular populations (Liu et al., 2019). Nanoproteomics has expanded the scope of turnover studies by enabling analysis of individual cells, laser-microdissected tissue regions, and spatially defined microenvironments. Applications include spatial turnover profiling within tumors (e.g., tumor core versus invasive margin), monitoring of rare circulating tumor cells, and longitudinal analysis of tissue-resident immune cells during disease progression or therapeutic intervention. These developments are driving the emergence of spatially resolved and temporally dynamic proteomics.

APPLICATIONS OF PROTEIN TURNOVER STUDIES

Biomarker discovery

Protein turnover kinetics provide a dynamic layer of biomarker information that often surpasses static protein abundance measurements in specificity and sensitivity. Proteins exhibiting altered rates of synthesis or degradation may more accurately reflect disease activity, progression, or therapeutic response (Table 1). In oncology, turnover-based biomarkers have the potential to predict treatment efficacy, emergence of resistance, and disease relapse at earlier stages than conventional biomarkers.

Therapeutic monitoring

Protein degradation pathways are central to the mechanisms of action of several modern therapeutic strategies, including proteasome inhibitors and proteolysis-targeting chimeras (Table 1). These therapies selectively induce degradation of disease-associated proteins, including oncogenic drivers and aggregation-prone proteins. Real-time monitoring of protein turnover enables direct evaluation of target engagement and degradation efficiency, providing a more accurate assessment of therapeutic activity than static quantification of protein levels alone (Ibrahim et al., 2025). Such dynamic measurements may support the optimization of drug dosing, early detection of therapeutic resistance, and development of personalized treatment strategies.

Personalized medicine

Protein turnover profiling offers a highly dynamic representation of patient-specific biology and has considerable potential in precision medicine. By quantifying protein synthesis and degradation rates, clinicians can identify subtle molecular differences among patients that are not detectable through genomic or transcriptomic analyses alone (Guo et al., 2025). At single-cell resolution, turnover analysis can reveal rare resistant subclones or cellular populations that evade therapy, thereby informing patient stratification and guiding tailored therapeutic interventions (Table 1). Integration of turnover profiling with multi-omics and spatial biology approaches is expected to play an increasingly important role in personalized diagnosis, prognosis, and treatment selection.

Tumor and immune heterogeneity

Single-cell turnover profiling has emerged as a powerful approach for understanding tumor heterogeneity and immune system dynamics. Within tumors, distinct cellular subpopulations frequently display unique turnover signatures associated with altered metabolism, proliferative capacity, metastatic potential, or resistance to therapy. Measuring protein synthesis and degradation at the single-cell level, therefore, provides insight into tumor evolution and adaptive resistance mechanisms. Similarly, immune cells undergo rapid proteomic remodeling during activation, differentiation, and pathogen response. Single-cell turnover

Table 1: Applications of single-cell proteomics.

	Application	Description / Purpose
1.	Cell type identification	Identifies unique proteomic signatures of different or rare cell types.
2.	Tumour heterogeneity	Analyses protein differences between cancer cells to understand tumor complexity.
3.	Immunology studies	Profiles immune cell subsets, their activation states, and signalling pathways.
4.	Developmental biology	Tracks protein changes during cell differentiation and embryonic development.
5.	Stem cell research	Studies pluripotency and lineage commitment through protein-level regulation.
6.	Neuroscience	Maps neuron subtypes and synaptic protein expressions in brain research.
7.	Tissue microenvironment	Dissects protein interactions in spatially organized tissues (e.g., tumors, brain).
8.	Biomarker discovery	Detects novel protein biomarkers for disease diagnosis and prognosis at the single-cell level.
9.	Infectious disease	Studies host-pathogen interaction at the cellular protein level.
10.	Ageing and senescence	Analysis of changes in protein expression in ageing cells or senescent states.
11.	Personalized medicine	Enables individualized therapy by profiling each patient's cell proteome.
12.	Drug response analysis	Access the individual cell response to drug treatment, essential for precision medicine.

approaches enable detailed investigation of dynamic proteomic changes in T cells, macrophages, dendritic cells, and other immune populations following infection, inflammation, or immunotherapy (Alam et al., 2025). Such analysis improves understanding of immune regulation and may facilitate the identification of biomarkers predictive of immune responsiveness and therapeutic outcomes

CHALLENGES AND FUTURE DIRECTIONS

The principal challenge is the intrinsic complexity and dynamic range of the proteome, particularly in clinical and heterogeneous biological samples where low-abundance proteins are often masked by highly abundant species (Jakobsen et al., 2021). Accurate quantification of protein synthesis and degradation rates also remains technically demanding because turnover kinetics are influenced by cell type, metabolic state, tissue microenvironment, and temporal fluctuations. *In vivo* studies introduce additional complexities, including incomplete isotope incorporation, precursor pool variability, and physiological perturbations caused by labelling strategies. At the analytical level, MS-based turnover experiments generate highly multidimensional datasets that require easy computational pipelines for peptide identification, isotopologue deconvolution, kinetic modelling, and statistical interpretation. Variability in sample preparation workflows, isotope-labelling protocols, instrument settings, and data-processing algorithms continues to limit reproducibility and cross-study comparability. Therefore, the development of standardized experimental guidelines, validated reference materials, and robust bioinformatics frameworks is essential for large-scale clinical implementation and regulatory acceptance (Mukherjee et al., 2025).

A major future direction is integrating protein turnover datasets with complementary multi-omics platforms, including transcriptomics, metabolomics, epigenomics, and spatial proteomics. Such integrative approaches will provide a systems-level understanding of cellular dynamics by linking transcriptional regulation, metabolic flux, and protein stability within defined cellular and tissue contexts. The emergence of spatially resolved and single-cell proteomics is particularly important, as these technologies enable direct investigation of cellular heterogeneity and microenvironment-specific proteome dynamics that are obscured in bulk analyses. Artificial intelligence and machine learning are expected to play transformative roles in the next generation of turnover studies. AI-driven models may improve peptide identification, predict protein half-lives, infer degradation pathways, and uncover previously unrecognized regulatory networks from large-scale dynamic proteomic datasets. Advances in live-cell imaging, multiplexed isotope tracing, and real-time MS are expected to enable direct visualization of protein synthesis and degradation with

unprecedented temporal and spatial resolution. Future translational applications will likely extend into precision medicine and digital pathology, where protein turnover signatures could serve as biomarkers for disease progression, therapeutic response, and patient stratification. Dynamic proteomics may ultimately support personalized treatment strategies by enabling real-time assessment of pathway activity and drug efficacy in individual patients.

CONCLUSION

Protein turnover maintains cellular homeostasis, proteostasis, and adaptation to physiological changes. Recent advances in stable isotope labelling, high-resolution mass spectrometry, computational modelling, and single-cell proteomics have transformed turnover analysis into a powerful systems biology tool for studying the dynamic regulation of the proteome. Unlike static protein measurements, turnover studies quantify protein synthesis and degradation rates, providing deeper insight into cellular function. These approaches have improved understanding of ageing, cancer, neurodegenerative and metabolic diseases, immune responses, tissue regeneration, etc. Emerging technologies, including spatial proteomics, AI-assisted analysis, and multi-omics integration, are expected to enhance clinical translation, precision medicine, and therapeutic development through detailed mapping of protein dynamics.

AUTHORS CONTRIBUTION

Conceptualization, writing-original draft preparation: P Y ; Review and editing: RS, SS; Visualization: MH; Conceptualization, editing and overall supervision: MKS.

DECLARATION OF INTERESTS

The authors declare no competing financial or commercial interests.

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