Computational tools to aid mass-resolved proteome measurements

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Abstract

Proteomic research gives important insight in biological functions and dynamics and as such may serve as an important tool in collecting molecular information for disease diagnostics. Liquid chromatography – mass spectrometry (LC-MS) has become indispensable tool in proteome exploration and allows the measurement of proteins, including modifications, potentially allowing early disease diagnosis and biomarker discovery. Due to the large complexity of proteomes, recognition of potentially important features may not always be easily perceived, therefore computational tools were developed that would allow the evaluation of specific area of LC-MS data, such that we can guide instrument operation based on the observable proteome.

In this study, proteins were analysed using bottom-up proteomics, in combination with the data independent acquisition (DIA) method named sequential windowed acquisition of all theoretical ion mass spectra (SWATH-MS). The SWATH-MS method iterates across the m/z range in wide m/z windows. Subsequently they are fragmented and measured as tandem mass spectra.

Several aspects of DIA and optimisation of this workflow have been evaluated. In particular mass-resolved (m/z) windows of proteomes were investigated. Traditional optimisation algorithms require several experiments and optimisation focusses on the total number of ions. In the work we reported here allows optimisation based on the highest coverage of proteins, and theoretically requires only a single measurement. The method developed was experimentally validated on human kidney tissue providing increased sensitivity and reproducibility to SWATH MS, compared to regular SWATH MS. The method can also provide short m/z windows in which proteins of interest lay, and thereby increase sensitivity on their peptide identifications/quantitation, in turn increase quantitation in proteins.