Data Independent Acquisition: Quo vadis?

Wednesday-10 workshop, June 3, 2020, 12:00-1:30 pm

68th Annual Meeting of the American Society of Mass Spectrometry (ASMS)

Organizers

a. Birgit Schilling, PhD
   Buck Institute for Research on Aging
b. Florian Meier, PhD
   Jena University Hospital and Max Planck Institute of Biochemistry

Invited Panelists

a. Ben C. Collins, PhD
   Queen's University Belfast
b. Yansheng Liu, PhD
   Yale School of Medicine
c. Lili Niu, M.Sc.
   Novo Nordisk Center for Protein Research, Copenhagen
d. Brian Searle, PhD
   Institute for Systems Biology, Seattle
e. Lynn Verbeke, M.Sc.
   Biognosys AG, Zurich

Introduction and Overview

Drs. Birgit Schilling (Buck Institute) and Florian Meier (Jena University Hospital) invited a panel of experts for research using data independent acquisition (DIA) to discuss current topics and future directions in the DIA workshop at ASMS Reboot 2020. Each of the panelists listed above represented one area of particular interest in this regard and, following an introduction round, each topic was discussed amongst the panelists and with the audience. To stimulate discussions, each panelist introduced his or her topic with a short overview of the state of the art and current challenges. Topics included spectral library generation, advances in library-free approaches, ion mobility spectrometry, cancer biology and signaling, as well as applications of DIA for clinical proteomics.

Panel Topics of Discussion

a. Library generation

Brian Searle discussed best practices for spectral library generation with a specific focus on Orbitrap mass spectrometers. The presentation highlighted the value of accurate retention times and chromatogram libraries, which can, for example, be efficiently

generated via gas phase fractionation. An interesting future direction is the combination with machine learning-based predictions.

b. Library-free DIA

Lynn Verbeke reported on recent developments in library-free approaches that overcome the necessity to generate experimental libraries. Computational advances now enable predictions of entire libraries *in silico* directly from FASTA files. However, in practice, applications thereof are limited due to computational demands and the extremely large search space. Library-free approaches, such as DIA-Umpire or directDIA, deconvolute complex DIA spectra into pseudo-DDA spectra, and now approach the performance of library-based DIA in certain experiments, thus presenting a very attractive strategy.

c. Ion mobility spectrometry

Ben Collins presented an overview of ion mobility-enhanced DIA methods, which add an additional dimension of separation to conventional LC-MS based methods. The technology is now widely commercially available and, most recently, encouraging proteomics results have been demonstrated with trapped ion mobility spectrometry (TIMS) and field-asymmetric ion mobility spectrometry (FAIMS) devices. Ion mobility technology holds promise to enable very fast analysis times down to minutes, but may also increase proteome coverage and increase sensitivity.

d. Cancer biology and signalling

Yansheng Liu reported on the application of DIA in cancer research, in particular the question how alterations in the genome, such as aneuploidy, affect protein copy numbers. An intriguing finding is that proteins involved in complexes may be buffered more efficiently than others. To study this in more detail and investigate proteome turnover on a whole-proteome level, Dr. Liu presented a novel workflow combining pulsed SILAC and DIA. This presentation also started a discussion on the different use cases of DIA and isobaric labeling techniques, such as TMT.

e. Clinical proteomics

Lili Niu provided an outlook on proteomics applications in a clinical setting. Potential applications include the discovery of biomarkers and drug targets, from body fluids, fresh tissue or archived material. The choice of mass spectrometry platforms and acquisition methods also depends on the study design, which could range from large scale studies with thousands of samples to N-of-1 studies. In particular for large studies, DIA can be advantageous. Future directions and challenges include analyses on the single cell level, proteoform analysis, quantifying post-translation modifications and integrating genomic information for personalized proteomics.