

Absolute Quantification of Peptides and Proteins: From Sample Preparation and Chromatography to ESI-MS/MS

Within the last decade, nanoscale liquid chromatography-tandem mass spectrometry (nLC-MS/MS) has revolutionized the identification of proteins and peptides in biological matrices. The technology has developed from being a purely qualitative tool for (global) proteomics to a quantitative method suitable for peptide/protein biomarker validation. Key to success has been the combination of highly specific sample preparation methods, high sensitivity nanospray ionization, and high performance mass spectrometry. In this short course, critical parameters involved in robust sample preparation, nanobore LC and nanospray along with their analytical benefits will be emphasized. The transition of the traditionally qualitative nLC-MS/MS technology to those suitable for absolute quantification will be discussed. Real world examples from the literature will be presented. The target audience includes both analysts that have been engaged in qualitative proteomics wishing to transition to quantitative analysis and small molecule quantitative analysts wishing to learn more about the challenges specific to peptide/protein quantification.

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