Methods, Challenges, and Goals in Isobaric Labeling Multiplexed Quantitative Proteomics

The massive increase in throughput of quantitative mass spectrometry-based proteomics through multiplexed data acquisition using isobaric labeling strategies, such as iTRAQ or TMT, has the potential to fundamentally change the type of questions we can ask about biological systems at the proteome level. Initial shortcomings of the technology regarding accuracy and precision of the quantitative results – the interference problem – were solved in our lab through the development of a gas phase purification strategy based on MS3 scans. Recent improvements of this method now allow the acquisition of protein expression profiles on a near-global scale. First applications in our lab include the study of the effect of deubiquitinating enzyme gene deletions on protein levels in yeast as well as the determination of protein abundance levels across multiple cancer cell lines. Another direction of our current research is to improve isobaric labeling multiplexed proteomics through extending the number samples that are quantified in a single experiment. A new generation of TMT reagents allows the parallel analysis of 18 samples.