

Quantitative Mass Spectrometry Analysis of the In Vivo Concentrations of Multiple, Pre-Selected Biomarker Proteins

Description

A technique called Multiple Reaction Monitoring scanning (MRM) on a triple quadrupole mass spectrometer (MS) is well known to be the most sensitive and specific way to validate novel candidate markers, protein isoforms, and post-translational modifications in a complex matrix such as plasma or a tissue extract. In a recent paper by Forest White and colleagues (Yadlin et. al., 2007), he compared MRM analysis to traditional discovery proteomics (IDA) in monitoring 222 peptides from complex samples. His results showed that reproducibility increased significantly from 34% (IDA) to 88% (MRM). In addition to the increased reproducibility of the MRM method, performing MRM-type analyses on a triple-quadrupole instrument also provides significant improvement in sensitivity and much greater dynamic range (five orders of magnitude). This is due to the MRM scan having extremely low noise because the mass spectrometer only transmits the parent ion of interest for fragmentation and then only detects a single daughter ion (very few co-eluting species meet this criteria). As a result of this, peptides can be detected at significantly lower concentrations, and maintain excellent S/N ratios. Using this MRM scan in conjunction with ultra high pressure chromatography systems (UPLC) and isotopically labeled idiotypic peptides, we plan to quantify the absolute levels of multiple, preselected proteins (e.g., up to 30 or more biomarker peptides/run) in a large number of samples from human sera/plasma, other biological fluids (e.g., CS fluid), and/or human cell/tissue extracts. Overall this technology should enable us to leverage the emerging mass spectrometry based paradigm for validating, quantitating, and potentially generating a new diagnostic tool for identification and confirmation of protein biomarkers from clinical samples.

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Multiple reaction monitoring for robust quantitative proteomic analysis of cellular signaling networks. Proc Natl Acad Sci U S A. 2007 Apr 3;104(14):5860-5. Epub 2007 Mar 26.