



**Identifying, Confirming, and Quantifying
Differentially Expressed Proteins in Rat Plasma:
A Workflow for Driving Biomarkers from Discovery to Clinical**

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Discovery and differential expression analysis are the key drivers for identifying potential biomarkers. However, the real bottleneck in biomarker research resides in developing validated methods in hopes of establishing clinical protocols. Problems associated with moving from discovery to clinical are attributed to correct identification of the peptide and ultimately, the protein to ensure the experimental results can be properly attributed to the proposed biomarker. To increase the efficiency in identifying potential biomarkers and developing validated methods, a high degree of confidence is critical for both stages. That is, correctly identifying the peptide sequence, ensuring specificity of the proteotypic peptide, and measurable response for a validation stage in the presence of a biological matrix for a targeted SRM analysis.

In this presentation, a workflow is discussed outlining biomarker identification using differential expression analysis on the LTQ Orbitrap XL and validating targeted SRM methods on a TSQ Quantum Ultra for control and diseased rat plasma. Key points in terms of confirming peptide and protein identification are discussed for both steps of the workflow as they relate to the experiment and interrogation of the data. In addition, new software advances designed to help expedite the workflow are discussed.