

## 137. Applications of a high-throughput targeted proteomics assay for analysis of engineered *Escherichia coli*

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### **Project Goals:**

Transformation of engineered *Escherichia coli* into a robust microbial factory is contingent on precise control of metabolism. Targeted proteomics is well suited for metabolic engineering due to rapid detection and accurate quantification of proteins. We created a high-throughput proteomics toolkit that provides a large number of validated and optimized targeted proteomics methods for absolute quantification of over 400 *E. coli* proteins. To facilitate high sample throughput, we have developed a fast chromatography method (10 minute total run time) and an automated sample preparation procedure that performs cell lysis, protein quantification, and tryptic digestion of hundreds of samples in a few hours. We demonstrate the utility of the toolkit by characterizing proteins in various metabolic pathways from exponentially growing *E. coli* cultured with glucose or xylose as the carbon source in aerobic and anaerobic conditions. From this work, principle component analysis using proteomics (PCAP) and correlation analysis methods that use targeted proteomic data have been developed to aid pathway optimization.