IPD Project Details

Project ID: IPD4939

Project Title: LC-MS/MS Proteomic Dataset of Klebsiella pneumoniae 4 clinical isolates Grown in Human Plasma and Mueller Hinton Broth at 0 and 2 Hours

Description: This dataset presents a comprehensive LC-MS/MS-based proteomic profile of Klebsiella pneumoniae 4 clinical isolates cultured under two distinct environmental conditions: human plasma and Mueller-Hinton Broth (MHB). The study design incorporates two early time points, 0 hours (baseline) and 2 hours post-incubation, to capture immediate and early-stage proteomic adaptations. These conditions were selected to enable direct comparison between a physiologically relevant host-mimicking environment and a standard nutrient-rich laboratory medium. The dataset comprises a total of 12 LC-MS/MS files, including 4 biological replicates at 0 hours in Mueller-Hinton Broth (MHB), 4 replicates at 2 hours in human plasma, and 4 replicates at 2 hours in MHB. Protein identification was performed using a reference Klebsiella pneumoniae proteome database from UniProt, and standard proteomics workflows were applied for peptide matching, false discovery rate (FDR) control, and protein inference.

Principal Investigator: Dr. Niraj Kumar

PI Affiliation: Associate Professor, Translational Health Science and Technology Institute (THSTI)

Sample Preparation: Cells collected from three bloodstream clinical isolates of K. pneumoniae grown in MHB and h-plasma for 2 hrs. were resuspended in 300 µL of urea CHAPS lysis buffer and kept on ice for 30 min, followed by sonication at 20% amplitude (6s on/6s off) for a total of 4 min on ice. The supernatant was collected, and cell debris was removed by centrifugation. Proteins were precipitated by the addition of chilled acetone and subsequently dissolved in 50 µL of 6 M urea prepared in a 50 mM ammonium bicarbonate buffer. For each condition, 40 µg of extracted protein was subjected to protein identification and quantification by mass spectrometry

Peptide Separation: Protein samples were digested using trypsin, allowing up to two missed cleavages. The resulting peptides were analyzed using LC–MS/MS on a TripleTOF 5600 mass spectrometer. No prior charge state deconvolution or deisotoping was performed, and no specific peptide enrichment strategy was applied. Tandem mass

spectra were acquired and searched against the Klebsiella pneumoniae FASTA database (306,428 entries) using Mascot (v2.6.2). Search parameters included a precursor ion tolerance of 0.30 Da and fragment ion tolerance of 0.30 Da. Carbamidomethylation of cysteine was set as a fixed modification, while oxidation of methionine and deamidation of asparagine and glutamine were considered variable modifications.

Protein Characterization: Protein identification and validation were performed using Scaffold (v4.11.0). Mascot search outputs were processed using PeptideProphet and ProteinProphet algorithms with delta-mass correction. Peptides were accepted at >50% probability, while proteins were accepted at >90% probability with at least one identified peptide. The false discovery rate (FDR) was ~3.6% for peptides and ~1.0% for proteins. Proteins sharing peptides were grouped based on parsimony principles. Quantitative analysis was performed using spectral counting within Scaffold with experiment-wide grouping. Protein annotations were mapped using UniProt/Swiss-Prot accession data, ensuring reliable and statistically robust protein identification across all samples.

Experiment Type: Bottom-up

Species: Klebsiella pneumoniae No Data

Tissue: Data in tissue_details No Data

Cell Type: Data in cell_details No Data

Disease: Data in disease_details No Data

Instrument Details: Data in instrument_details Data in instrument_details

Protein Modifications:

PubMed ID: