

Mass Spectral Proteomics Methods and Data Check list

This checklist is intended to help authors submit manuscripts that present mass spectral proteomics methods, data and figures in a transparent manner that is compatible with JBC guidelines and best practices in the field, and facilitates reproducing the study. More detailed instructions are available at: [Molecular and Cellular Proteomics Guidelines](#).

A. Source and Isolation of Biological Samples

Organism (human or other)

Number of technical replicates

Number of biological replicates

Cell/tissue lysis and fractionation conditions, including how samples were protected from proteolysis, dephosphorylation etc.

Protein or peptide enrichment conditions (i.e. immunoprecipitation, affinity methods for PTMs (i.e. phosphorylation or other)

Proteolytic digestion conditions, time, temperature, protease type and specificity

Peptide clean up and off-line chromatography methods

B. Mass Spectrometry (MS) Conditions (LC-MS or MALDI-TOF-MS)

Instrument name (vendor/model), including HPLC and mass spectrometer

Details on HPLC column(s), description of mobile phases, flow rate, gradients etc.

MS parameters including detailed conditions for data-dependent analysis (DDA), data-independent analysis (DIA) or MALDI MALDI-TOF-MS/MS applications. Including all MS1 and MS2 (or higher-MS3) parameters, resolution, instrument settings etc. For example, included MS/MS scans resolution and dynamic exclusion conditions, collision energy and type.

Methods of peptide/protein quantification (i.e. TMT, ITRAQ, SILAC or other) or label -free quantification (i.e. MaxQuant).

C. Data analysis and processing methods

Name of software and methods for generating peak lists.

Database searching software and version/date

Peptide exclusion conditions, number of unique and razor peptides

Database(s) used including name, version and taxonomy

Fixed and variable modifications, number of missed cleavages allowed, precursor and product ion mass tolerances

Calculation of false discovery rate (FDR) and criteria for acceptant of peptide and protein assignments (i.e. Benjamini–Hochberg method)

D. Statistics and data validation

Type of software or name of program used, including version

Data clustering conditions (if applicable)

Citation of literatures sources used

Methods to determine significance, power analysis or test(s) applied (i.e. modified T-tests, etc.)

E. Data reporting

A supplementary file in Excel format is recommended, which should include:

Protein data

Should include protein name, accession number and molecular weight.

Number of assigned spectra and unique sequences used for protein identification.

Percent sequence coverage, database search score

Peptide data

Include peptide sequence data with start and stop residue numbers, observed mass, mass error. Scores/expect values for assignment, post-translational modifications, probability determination for modification sites

The complete MS proteomics data set must be deposited to a publicly available database (e.g. ProteomeXchange Consortium via the PRIDE partner repository, MASSive); details of how to access the data must be provided in the manuscript.

F. Predictions from Proteomics Data and presentation

Pathway analysis platforms used (i.e. Ingenuity Pathway Analysis (IPA), RoKAI, Kinase-library.phosphosite.org others).

Generation of kinome pathways (Coral, Kinome Render)