

Proteomics Informatics Spring 2022 (BMSC-GA 4437)

Lecturers:

David Fenyo (David@FenyoLab.org), Professor

Beatrix Ueberheide (Beatrix.Ueberheide@nyumc.org), Associate Professor

Lizabeth Katsnelson (Lizabeth.Katsnelson@nyulangone.org), PhD Student

Course overview

This course will give an introduction of proteomics and mass spectrometry workflows, experimental design, and data analysis with a focus on algorithms for extracting information from experimental data. The following subjects will be covered in: (1) Protein identification (peptide mass fingerprinting, tandem mass spectrometry, database searching, spectrum library searching, de novo sequencing, significance testing); (2) Protein characterization (protein coverage, top-down proteomics, post-translational modifications, protein processing and degradation, protein complexes); (3) Protein quantitation (metabolic labeling - SILAC, chemical labeling, label-free quantitation, spectrum counting, stoichiometry, biomarker discovery and verification). Examples will be provided throughout the course on how the different approaches can be applied to investigate biological systems. The class will be structured to include hands-on practical techniques for analyzing relevant proteomics datasets.

Learning objectives

At the conclusion of the course, the student will be able to:

Understand experimental design for mass spectrometry based proteomics;

Demonstrate detailed understanding of the possibilities and limitations of algorithms that are applied to proteomics data; and

Analyze a large proteomics data set using available algorithms.

Course Assessment

Readings and participation (30%): Students are required to attend class, to complete reading assignments and to participate in discussions and engage in healthy exchange of ideas. Each student is required to lead at least one reading from the assigned weekly readings. This discussion lead will be graded.

Assignments (30%): Programming assignment will be given at the end of each class, and the solutions to these assignments should be e-mailed to Assignments@FenyoLab.org within a week.

Final project (40%)

Missed Exams and Grade Appeals

Make-up examinations (for final only) will be given under special circumstances. Documentation will be required to verify a student's claim. If a make-up exam is permitted, a different exam will be written for that student and may have a different format than the regular examination.

The assignments must be turned in on time and no late assignments will be accepted.

If there is a time that you believe that there is a mistake in grading of an assignment/exam, you will have a chance to appeal your exam grade within a week after you receive your grade. If you think this is the case, you must write a note describing the error, attach it to the original exam, and give it to me within a week of the return of your exam. I will review your argument and my initial grading, and then return your exam with a decision to you in a timely manner.

General Policies

Late/missed work: You must adhere to the due dates for all required submissions. If you miss a deadline, then you will not get credit for that assignment/post. Try to avoid last minute submissions.

Incompletes: No "Incompletes" will be assigned for this course unless we are at the very end of the course and you have an emergency.

Responding to Messages: I will check e-mails daily during the week, and I will respond to course related questions within 48 hours.

Announcements: I will make announcements throughout the semester by e-mail. Make sure that your email address is updated; otherwise you may miss important emails from me.

Safeguards: Always back up your work on a safe place (electronic file with a backup is recommended) and make a hard copy. Do not wait for the last minute to do your work. Allow time for deadlines.

Plagiarism: Plagiarism, the presentation of someone else's words or ideas as your own, is a serious offense and will not be tolerated in this class. The first time you plagiarize someone else's work, you will receive a zero for that assignment. The second time you plagiarize, you will fail the course with a notation of academic dishonesty on your official record.

Lecture 1 Introduction: Proteomics (January 27, 2022 at 5:30pm)

Lecturer: Fenyo

Background Reading

A. Bensimon, A.J.R. Heck R. Aebersold, "Mass Spectrometry–Based Proteomics and Network Biology", Annual Review of Biochemistry 81 (2012) 379-405.

Lundberg E, Borner GHH. Spatial proteomics: a powerful discovery tool for cell biology. Nat Rev Mol Cell Biol. 2019

Lecture 2 Interpretation of mass spectra & denovo sequencing I (February 3, 2022 at 5:30pm)

Lecturer: Ueberheide

Background Reading

Seidler J, Zinn N, Boehm ME, Lehmann WD, "De novo sequencing of peptides by MS/MS, Proteomics 10 (2010) 634-49.

Standing KG, "Peptide and protein de novo sequencing by mass spectrometry, Curr Opin Struct Biol. 13 (2003) 595-601.

Lecture 3 Interpretation of mass spectra & denovo sequencing II (February 10, 2022 at 5:30pm)
Lecturer: Ueberheide

Lecture 4 Protein identification (February 17, 2022 at 5:30pm)
Lecturers: Fenyo and Katsnelson

Background Reading

D. Fenyo, J. Eriksson, R. Beavis, "Mass spectrometric protein identification using the global proteome machine", Methods Mol Biol 673 (2010) 189-202.

GPMDDB Tutorials

Discussion Reading

Elias, J.E. & Gygi, S.P.: "Target-decoy search strategy for increased confidence in large-scale protein identifications by mass spectrometry" Nat. Methods 4, 207-214 (2007).

Gilbert S. Omenn: Reflections on the HUPO Human Proteome Project, the Flagship Project of the Human Proteome Organization, at 10 Years, Molecular & Cellular Proteomics 20100062

Lecture 5 Protein quantitation I (February 24, 2022 at 5:30pm)
Lecturer: Fenyo

Background Reading

M.A. Gillette, S.A. Carr, "Quantitative analysis of peptides and proteins in biomedicine by targeted mass spectrometry", Nature Methods 10 (2013) 28-34.

D.R. Mani, S.E. Abbatiello, S.A. Carr, "Statistical characterization of multiple-reaction monitoring mass spectrometry (MRM-MS) assays for quantitative proteomics", BMC Bioinformatics 13 (2012) S9.

Discussion Reading

Domon & Aebersold: Options and considerations when selecting a quantitative proteomics strategy, Nat. Biotechnol 28, 710-721 (2010).

Shenoy et al.: Proteomic patterns associated with response to breast cancer neoadjuvant treatment, Molecular Systems Biology (2020)16:e9443.

Lecture 6 Proteogenomics (March 3, 2022 at 5:30pm)
Lecturer: Fenyo

Background Reading

Ruggles KV, Krug K, Wang X, Clauser KR, Wang J, Payne SH, Fenyő D, Zhang B, Mani DR. Methods, Tools and Current Perspectives in Proteogenomics. Mol Cell Proteomics 2017

Discussion Reading

Dou et al.: Proteogenomic Characterization of Endometrial Carcinoma, Cell 180 (2020) 729-748.e26

Gillette et al.: Proteogenomic Characterization Reveals Therapeutic Vulnerabilities in Lung Adenocarcinoma. Cell 182 (2020) 200-225.e35

Lecture 7 Protein quantitation II (March 10, 2022 at 5:30pm)

Lecturer: Katsnelson

Background Reading

Tyanova, S., Temu, T., and Cox, J., The MaxQuant computational platform for mass spectrometry-based shotgun proteomics, Nat Protocols, 2016, 11, pp 2301–2319

Pino LK et al., The Skyline ecosystem: Informatics for quantitative mass spectrometry proteomics. Mass Spectrom Rev. 2017

Spring Break March 14-20, 2022

Lecture 8 Protein characterization: post-translational modifications (March 24, 2022 at 5:30pm)

Lecturer: Ueberheide

Background Reading

Protein characterization I: PTMs

Discussion Reading

Leutert et al.: Decoding Post-Translational Modification Crosstalk With Proteomics, Molecular & Cellular Proteomics, Volume 20, 100129

Lu et al.: Accelerating the Field of Epigenetic Histone Modification Through Mass Spectrometry–Based Approaches, Molecular & Cellular Proteomics, Volume 20, 100006

Discussion Reading

Leutert et al.: Decoding Post-Translational Modification Crosstalk With Proteomics, Molecular & Cellular Proteomics, Volume 20, 100129

Lecture 9 Metabolomics (March 31, 2022 at 5:30pm)

Lecturer: Jones

Background Reading

Misra: New software tools, databases, and resources in metabolomics: updates from 2020. *Metabolomics*. 2021 May 11;17(5):49. doi: 10.1007/s11306-021-01796-1. PMID: 33977389; PMCID: PMC8112213.

Alseekh et al.: Mass spectrometry-based metabolomics: a guide for annotation, quantification and best reporting practices. *Nat Methods*. 2021 Jul;18(7):747-756. doi: 10.1038/s41592-021-01197-1. Epub 2021 Jul 8. PMID: 34239102; PMCID: PMC8592384.

Discussion Reading

Li et al.: Spectral entropy outperforms MS/MS dot product similarity for small-molecule compound identification. *Nat Methods*. 2021 Dec;18(12):1524-1531. doi: 10.1038/s41592-021-01331-z.

Lecture 10 Project Presentations and Paper Discussions (April 7, 2022 at 5:30pm)

Discussion Reading

Schmid et al.: Ion identity molecular networking for mass spectrometry-based metabolomics in the GNPS environment. *Nat Commun*. 2021 Jun 22;12(1):3832. doi: 10.1038/s41467-021-23953-9.

Petelski et al: Multiplexed single-cell proteomics using SCoPE2, *Nat Protoc*. 16 (2021) 5398-5425.

Lun, Xiao-Kang et al.: Profiling Cell Signaling Networks at Single-cell Resolution, *Molecular & Cellular Proteomics*, Volume 19, Issue 5, 744 - 756

Lu et al.: Accelerating the Field of Epigenetic Histone Modification Through Mass Spectrometry–Based Approaches, *Molecular & Cellular Proteomics*, Volume 20, 100006

Lecture 11 Protein characterization: protein interactions 1 (April 14, 2022 at 5:30pm)

Lecturer: Ueberheide

Background Reading

Protein characterization II: protein interactions

Z Hakhverdyan, M Domanski, LE Hough, AA Oroskar, AR Oroskar, S Keegan, DJ Dilworth, KR Molloy, V Sherman, JD Aitchison, D Fenyő, BT Chait, TH Jensen, MP Rout, J LaCava, "Rapid, optimized interactomic screening", *Nature Methods* 2015

A Leitner, R Reischl, T Walzthoeni, F Herzog, S Bohn, F Förster, and R Aebersold, "Expanding the Chemical Cross-Linking Toolbox by the Use of Multiple Proteases and Enrichment by Size Exclusion Chromatography", *Mol Cell Proteomics* 11 (2012)

Discussion Reading

Sinz: Cross-Linking/Mass Spectrometry for Studying Protein Structures and Protein–Protein Interactions: Where Are We Now and Where Should We Go from Here? 57 (2018) 6390-6396

Salas, Daniela et al.: Next-generation Interactomics: Considerations for the Use of Co-elution to Measure Protein Interaction Networks, *Molecular & Cellular Proteomics*, Volume 19, Issue 1, 1 - 10

Lecture 12 Protein characterization: MHC Peptides (April 21, 2022 at 5:30pm)
Lecturer: Katsnelson

Background Reading

Purcell AW, Ramarathinam SH, Ternet N. Mass spectrometry-based identification of MHC-bound peptides for immunopeptidomics. *Nat Protoc.* 2019 Jun;14(6):1687-1707.

Freudenmann LK, Marcu A, Stevanovic S. Mapping the tumour human leukocyte antigen (HLA) ligandome by mass spectrometry. *Immunology.* 2018 Jul;154(3):331-345.

Discussion Reading

Qi et al.: Proteogenomic Analysis Unveils the HLA Class I-Presented Immunopeptidome in Melanoma and EGFR-Mutant Lung Adenocarcinoma. *Mol Cell Proteomics.* 2021;20:100136.

Weingarten-Gabbay et al.: Profiling SARS-CoV-2 HLA-I peptidome reveals T cell epitopes from out-of-frame ORFs. *Cell.* 2021 Jul 22;184(15):3962-3980.e17.

Lecture 13 Single-Cell Proteomics (April 28, 2022 at 5:30pm)
Lecturer: Loomis

Background Reading

Slavov: Scaling Up Single-Cell Proteomics 21 (2021) 100179.

<https://doi.org/10.1016/j.mcpro.2021.100179>

Schürch et al.: Coordinated Cellular Neighborhoods Orchestrate Antitumoral Immunity at the Colorectal Cancer Invasive Front. *Cell.* 2020 Oct 29;183(3):838. doi: 10.1016/j.cell.2020.10.021.
Erratum for: *Cell.* 2020 Sep 3;182(5):1341-1359.e19.

Lecture 14 Proteomics Tools for Characterizing Macromolecular Assemblies (May 5, 2022 at 5:30pm)
Lecturer: Chait

Background Reading

B.T. Chait, M. Cadene, P.D. Olinares, M.P. Rout, Y. Shi. "Revealing Higher Order Protein Structure Using Mass Spectrometry", *J Am Soc Mass Spectrom.* 27 (2016) 952-965

Lecture 15 Project Presentations (May 12, 2022 at 5:30pm)