Master 2 research internship in Integrated Structural & Cell Biology in Grenoble

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Host laboratory:
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Host group/team:
EDyP

Title of the M2 research internship:
Deciphering the detailed composition of intact nucleosomes by mass spectrometry

Project summary:
Nucleosomes constitute the structural units which allow 3D organization of DNA and modulate access to the genome to achieve gene transcription, repair and duplication. Nucleosomes are made up of two copies of each of the core histones H2A, H2B, H3 and H4. The composition of nucleosomes in terms of histone variant sequences and multiple post-translational modifications (PTMs) on lysine, arginine and serine residues exquisitely regulate all DNA-templated processes. For example, variant H3.3 and lysine acetylation on multiple residues are usually associated with active transcription. The information of histone variant and PTM relative amounts can be provided by mass spectromety (MS) analysis of histones, classically after their enzymatic proteolysis to produce peptides of 10-40 amino acids. This type of analysis provides valuable insights into the changes in relative abundances of (variant x PTM) combinations during cell differentiation processes or between healthy and diseased tissues. However, the co-existence of PTMs in a given histone molecule and the preferential association of PTMs and histone variants in the same nucleosome cannot be grasped by these analyses. To decipher the histone code orchestrated within nucleosomes, an analytical method which maintains intact these structures is required. In 2021, such a proteomic approach named Nuc-MS was published, which makes the most of a mass spectrometer equipped with a high-mass detection range and a fragmentation mode well suited to the analysis of high mass and charge peptides (Electron Transfer Dissociation). Our lab has recently acquired such an instrument. In this project, we aim to build on our expertise in mass spectrometry and analysis of protelyzed histones to establish this cutting-edge analytical approach. We will apply it on commercial nucleosomes, nucleosomes extracted from mouse brain (WT and a model for a neurodegenerative disease) and the subset of nucleosomes enriched by immuno-precipitation of histone H3 bearing a specific acetylation mark.

Keywords:
nucleosomes, proteomic,

Relevant publications of the team:

Neutral mass spectrometry of virus capsids above 100 megadaltons with nanomechanical resonators.

Sergio Dominguez-Medina, Shawn Fostner, Martial Defoort, Marc Sansa, Ann-Kathrin Stark, Mohammad Abdul Halim, Emeline Vernhes, Marc Gely, Guillaume Jourdan, Thomas Alava, Pascale Boulanger, Christophe Masselon, Sébastien Hentz.

Science. 2018 Nov 23;362(6417):918-922. doi: 10.1126/science.aat6457.

Proteoform: a single term describing protein complexity.

[...] Christophe Masselon, Michael Gross, Fred McLafferty, Yury Tsybin, Ying Ge, Ian Sanders, James Langridge, Julian Whitelegge, Alan Marshall (Consortium for Top Down Proteomics).

Nat Methods. 2013 Mar;10(3):186-7. doi: 10.1038/nmeth.2369.

Multi-omic analysis of gametogenesis reveals a novel signature at the promoters and distal enhancers of active genes.

Crespo M, Damont A, Blanco M, Lastrucci E, El Kennani S, [...], Govin J, Fenaille F, Battail C, Cocquet J, Pflieger D.

Nucleic Acids Res. 2020 May 7;48(8):4115-4138. doi: 10.1093/nar/gkaa163.

Small Mass but Strong Information: Diagnostic Ions Provide Crucial Clues to Correctly Identify Histone Lysine Modifications.

Hseiky A, Crespo M, Kieffer-Jaquinod S, Fenaille F, Pflieger D.

Proteomes. 2021 Apr 23;9(2):18. doi: 10.3390/proteomes9020018.

Systematic quantitative analysis of H2A and H2B variants by targeted proteomics.

El Kennani S, Adrait A, Permiakova O, Hesse AM, Ialy-Radio C, Ferro M, Brun V, Cocquet J, Govin J, Pflieger D.

Epigenetics Chromatin. 2018 Jan 12;11(1):2. doi: 10.1186/s13072-017-0172-y.