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

Supervisor(s):
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Host laboratory:
BGE bge-lab.fr/

Host group/team:
Gen&Chem

Title of the M2 research internship:

Exploring the role of ubiquitin proteases in transcriptional regulation using an innovative genetic screen

Project summary:

Gene transcription is an integration point for numerous stimuli, it conditions its normal or pathological development. Like most cellular functions, transcription is regulated by the ubiquitin system, which consists of the reversible addition of ubiquitin groups to proteins and can modulate their properties. This system is orchestrated by the antagonistic activities of more than 600 ubiquitin ligases and 100 deubiquitinases (DUBs). DUBs are of particular interest as a source of therapeutic targets: their deregulation is characteristic of certain cancers and they can be chemically inhibited. However, the ubiquitin system is complex, involves interconnected signalling mechanisms, with numerous substrates and partners per DUB. Yet, the specific impact of each DUB is challenging to decipher.

The general aim of the project is to identify DUBs with a direct role in transcription and characterise their modes of action, in human cells. Ultimately, an original large-scale genetic screen with high temporal resolutions will be implemented. The specific goal of this M2 internship is to set up the molecular and cellular tools necessary for this genetic screen. The student will work on 2-3 selected DUB genes (such as USP36, USP7, BAP1, previously involved in transcriptional regulation) and generate cells to achieve their rapid and inducible repression, using a recently developed CRISPR/Cas9-based system. This includes design and cloning of guide RNAs, establishment of cell lines, validation and optimisation of the system. In parallel, the student will implement a method to monitor global levels of transcription in a cell, by imaging the incorporation of a uridine analogue into nascent RNA transcripts.

This internship is a pilot project, as part of the launch of a new research theme in the laboratory. The results generated will serve as a foundation to implement a screen applied to all nuclear DUBs. The student will be exposed to a variety of technologies, building on the team's expertise in molecular engineering, cellular imaging, DUBs and transcription.

Keywords:

transcription, ubiquitin signaling, molecular and cellular engineering

Relevant publications of the team:

- Milligan L, SAYOU C, Tuck A, Auchynnikava T, Reid JEA, Alexander R, De Lima Alves F, Allshire R, Spanos C, Rappsilber J, Beggs JD, Kudla G, Tollervey D. RNA polymerase II stalling at pre-mRNA splice sites is enforced by ubiquitination of the catalytic subunit. eLife (2017); 10.7554/ELIFE.27082.

- SAYOU C, Millán-Zambrano G, Santos-Rosa H, Petfalski E, Robson S, Houseley J, Kouzarides T, Tollervey D. RNA binding by the histone methyltransferases Set1 and Set2. Molecular and Cellular Biology (2017); 10.1128/MCB.00165-17.

- Cappuccio G*, SAYOU C*, Le Tanno P*, [...], Thevenon J, Govin J, Vitobello A, Brunetti-Pierri N. De novo SMARCA2 variants clustered outside the helicase domain cause a new recognizable syndrome with intellectual disability and blepharophimosis distinct from Nicolaides-Baraitser syndrome. Genetics in Medecine (2020); 10.1038/S41436-020-0898-Y.

- Franco G, TAILLEBOURG E, Delfino E, Homolka D, Gueguen N, Brasset E, Pandey RR, Pillai RS, FAUVARQUE MO. The catalytic-dead Pcif1 regulates gene expression and fertility in Drosophila. RNA (2023) 10.1261/rna.079192.122.

- Thevenon D, Seffouh I, Pillet C, Crespo-Yanez X, FAUVARQUE MO, TAILLEBOURG E. A Nucleolar Isoform of the Drosophila Ubiquitin Specific Protease dUSP36 Regulates MYC-Dependent Cell Growth. Front Cell Dev Biol. (2020); 10.3389/fcell.2020.00506.

- Crespo-Yàñez X, Aguilar-Gurrieri C, Jacomin AC, Journet A, MORTIER M, TAILLEBOURG E, SOLEILHAC E, Weissenhorn W, FAUVARQUE MO. CHMP1B is a target of USP8/UBPY regulated by ubiquitin during endocytosis. PLoS Genet. (2018); 10.1371/journal.pgen.1007456.