EuReCa International PhD Program

PhD thesis project

2021 Call for application

Novel chemical tools to study base-excision DNA repair

General information

Call	2021
Reference	2021-06-GRANZHAN_BOMBARD
Keyword(s)	Chemical biology; fluorescent probes; DNA damage; DNA probes; DNA sequencing

Director(s) and team

Thesis director(s)	Anton Granzhan & Sophie Bombard
Research team	Chemistry for Nucleic Acid Recognition
Research department	UMR9187 / U1196 - Chemistry and Modelling for the Biology of Cancer (CMBC)

Description of the PhD thesis project

Apurinic/apyrimidinic (AP, or abasic) sites represent key intermediates in base excision DNA repair (BER), the repair pathway responsible for the maintenance of genome integrity through removal of oxidative and alkylated DNA damage. Quantification of the global level and genomic mapping of AP sites are crucial for understanding the distribution of different types of DNA damage (oxidized, alkylated or deaminated bases), both endogenous and induced by anti-cancer drugs.

The available methods for the detection of AP sites rely on the reaction of the open (aldehyde) form of AP sites with aldehyde-reactive chemical probes possessing an oxyamine or a hydrazine group; as a consequence, the reactivity of these probes with 5-formylcytosine and 5-formyluracil, naturally present in the genome, is a serious issue which requires sophisticated protocols for avoiding cross-reactivity and can lead to biased results.

Along these lines, we have previously developed a family of macrocyclic ligands, based on the bis-naphthalene (“BisNP”) scaffold, that strongly and selectively bind to AP sites in DNA with nanomolar affinity and, depending on the substitution pattern, induce DNA strand cleavage at AP sites, or generate chemically stable covalent ligand–DNA adducts through a mechanism different from other aldehyde-reactive probes (Caron et al., Chem. Eur. J. 2019, 25, 1949).

In this project, we will exploit this scaffold to develop novel chemical tools to evaluate the global level of AP sites in genomic DNA and to map them in the genome.
Specifically, (i) non-covalent probes will be designed to quantify the global level of AP sites through fluorescence measurements, whereas (ii) covalent probes will be developed to capture and map the distribution of AP sites in the genome through DNA sequencing. The utility of these probes will be validated in cells treated with DNA-damaging drugs.

International, interdisciplinary & intersectoral aspects of the project

This project spans across several disciplines, namely organic chemistry (synthesis and characterization of the probes), molecular biophysics and biochemistry (probe–DNA interaction studies) and cellular biology (cell cultures).

The methods for quantification and genomic mapping of AP sites developed within the framework of this project will be protected through patent applications, if applicable.

Parts of the project, in particular the development of protocols for isolation of AP-DNA from cells, or in-cell studies of binding of probes to AP-DNA, will be performed in collaboration with labs abroad (USA, Czech Republic).

Recent publications

1. Duskova K, Lejault P, Benchimol É, Guillot R, Britton S, **Granzhan A**, Monchaud D. DNA junction ligands trigger DNA damage and are synthetic lethal with DNA repair inhibitors in cancer cells. J. Am. Chem. Soc. 2020, 142, 424–435. https://doi.org/10.1021/jacs.9b11150

2. Caron C, Duong XNT, Guillot R, Bombard S, **Granzhan A**. Interaction of Functionalized Naphthalenophanes with Abasic Sites in DNA: DNA Cleavage, DNA Cleavage Inhibition, and Formation of Ligand–DNA Adducts. Chem. Eur. J. 2019, 25, 1949–1962. https:// doi.org/10.1002/chem.201805555.

3. Krafcikova M, Dzatko S, Caron C, **Granzhan A**, Fiala R, Loja T, Teulade-Fichou MP, Fessl T, Hänsel-Hertsch R, Mergny JL, Foldynova-Trantirkova S, Trantirek L. Monitoring DNA–ligand interactions in living human cells using NMR spectroscopy. J. Am. Chem. Soc. 2019, 141, 13281–13285. https://dx.doi.org/10.1021/jacs.9b03031

4. Saha A, Bombard S, **Granzhan A**, Teulade-Fichou MP. Probing of G-quadruplex structures via ligand-sensitized photochemical reactions in BrU-substituted DNA. Sci. Rep. 2018, 8, 15814. https://dx.doi.org/10.1038/s41598-018-34141-z

5. Kotera N, Poyer F, **Granzhan A**, Teulade-Fichou MP. Efficient inhibition of human AP endonuclease 1 (APE1) via substrate masking by abasic site-binding macrocyclic ligands. Chem. Commun. 2015, 51, 15948–15951. https://doi.org/10.1039/C5CC06084B

Expected profile of the candidate

Applicants should hold a Master's degree in organic or bioorganic chemistry (or equivalent) and be motivated to carry out a multi-disciplinary research project at the interface of chemistry and biology.

It is strongly recommended to have solid, proven skills in organic chemistry and usual analytical methods (NMR, LC/MS, HPLC). Knowledge and hands-on experience with biochemistry and molecular biology techniques and/or cell cultures would be a strong advantage.

Candidates holding a Master's degree in Biochemistry and having hands-on experience in organic synthesis are also encouraged to apply.