Ecole Doctorale COMPLEXITE DU VIVANT – Fiche Projet CONCOURS

Fiche à nommer selon le format Nom_Prenom (sans accents ni cédilles), à enregistrer en format PDF et à renvoyer à l'adresse : edcdv@sorbonne-universite.fr

Nom et prénom du directeur de thèse (et si besoin du co-directeur) : Marbouty, Martial
Le directeur de thèse et le co-directeur doivent impérativement avoir l'HDR ou équivalent
Coordonnées Tel : 0145688016 e-mail : martial.marbouty@pasteur.fr

Nom et prénom du co-encadrant: Espeli, Olivier (HDR, 2009)
Coordonnées Tel :0144271249
e-mail :olivier.espeli@college-de-france.fr

Y-a-t-il un candidat déjà identifié pour le projet: OUI ~~NON~~

Nom et prénom du responsable de l'équipe : Koszul Romain

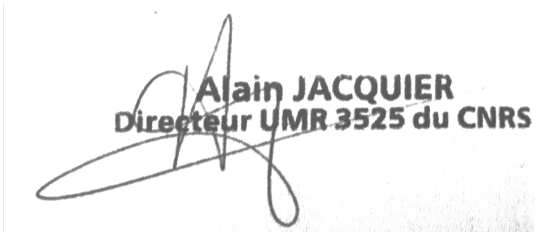
Intitulé de l'équipe : Régulation Spatiale des Génomes
Nombre de chercheurs et enseignants-chercheurs statutaires de l'équipe titulaires d'une HDR (ou équivalent) : 2 + dérogation Cournac (ANR JCJC, HDR prévue 2022)

Nom et prénom du responsable d'UMR ou de département: Jacquier, Alain

Intitulé et N° d'UMR ou de département : UMR 3525

Titre du projet de thèse *: analysis of the mecanisms of bacteriophage T4 Ndd protein on the disruption of Escherichia coli nucleoid*

Signature du directeur d'UMR ou de département (vaut avis favorable pour le dépôt du projet) :



Spécialité : microbiologie, génomique

Résumé du projet de thèse *(1 page maximum, en anglais)*
Pour les thèses avec 2 co-directeurs, ou en partenariat entre 2 laboratoires ou structures, indiquer la participation de chaque co-directeur et structure dans la gestion du projet.

Bacterial chromosomes are gigantic DNA molecules packed into a small cellular subcompartment called the nucleoid. Using a wide range of techniques, co-supervisors hold a strong track record of leading studies that have helped determine how different protein factors contribute to the organization of this structure. Bacteriophages, viruses of bacteria, have coevolved with bacteria for eons and have consequently evolved mechanisms to specifically and optimally inhibit or hijack key metabolic functions of their bacterial host. The T4 phage is an obligate lytic phage of E. coli, whose successful growth cycle results in host lysis. This model bacteriaophage was an important player in the history of genetics and molecular biology and, as such, has been the subject of intensive research for 80 years. Specific functions have been assigned to many gene products of the phage genome. For instance, the structural elements responsible for the capside assembly of the phage are well characterized, as are gene products that are involved in the replication of the phage DNA. However, despite intensive study, the mechanism by which it manipulates host functions remains unclear.

A series of studies have shown that the expression of only one of the T4 genome gene, the Nucleoid Disorganization Deficient (Ndd) gene, is notably sufficient to induce the disruption of the nucleoid, followed and subsequent death of the bacteria. Indeed, this small protein alone induces a relocalization of the bacterial nucleoid to the cell periphery within a few minutes. The molecular pathways behind this intriguing but striking phenomena have remained for the large part unexplained until today, despite the fundamental as well as applied interest that it could have. We hypothesized that this phenomenon could also reveal structural characteristics of the bacterial nucleoid, a research theme of our two laboratories.

The team of MM at the Institut Pasteur has tackled the challenge to revisit, using state-of-the-art molecular tools, this phenomena. A student in the laboratory developed an amenable, inducible system that can easily be manipulated and tested using imaging (O. Espeli Team) and "omics" approaches (M. Marbouty Team). This allowed us to investigate the changes in the organization over time of the nucleoid using "chromosome conformation capture" (3C or Hi-C) transcriptomic and microscopic approaches in cells expressing Ndd. These experiments confirmed some of the observations made in former works, but also showed that Ndd expression induces the formation of localized DNA breaks and induces DNA repair pathways. In addition, we made intriguing observation pointing at a putative association of the Ndd protein directly to its locus, which may hinder replication and promote DNA damages. However, these data did not allow to identify precisely the molecular mechanism of action of Ndd.

The PhD project aims at enhancing the characterization of Ndd functioning and solve its mechanisms of action. First, we will attempt to fish out putative Ndd targets on the genome and proteome of E. coli respectively with Chromatin immunoprecipitation and Co-IP experiments. Second, we will monitor Ndd activity in the context of a complete T4 phage infection. Third, we will characterize the respective impact of having Ndd carried on a plasmid, or on a chromosome, to decipher whether Ndd binds directly to DNA in the immediate vicinity of its transcription unit. Finally, we will search for partners of Ndd, among bacterial and phage genes, and see if we can mobilize them to develop an active Ndd mechanism in another species: first, in a closely related bacterium, and then in more distant species such as the budding yeast *Saccharomyces cerevisiae*. We will follow nucleoid disruption by live microscopy and by performing time series Hi-C and transcriptomic experiments. Increasing knowledge of the bacterial pathways targeted by Ndd and identifying the disruptive gene products used against them may generate tools to manipulate the E. coli genome.

Faisabilité du projet de thèse (*1/2 page maximum, en anglais)*

Expliciter la faisabilité du projet en terme d'expertise de l'équipe d'accueil, des collaborations potentielles qui pourront être mises en place pour certains aspects du projet, de la disponibilité des appareils nécessaires au bon déroulement du projet...

The PhD student will work in the highly interdisciplinary environment of the RSG team, encompassing ~10 people (3 permanent researchers, 1 permanent IP engineer, 4 PhD, 3 postdocs). The interdisciplinary nature of the projects results from collaboration between microbiologists, geneticists, physicists and computer scientists, either in the lab or through collaborations. Although, Phage biology is an emerging topic lead by Martial Marbouty in the RSG group. We recently developed a meta-HiC approach to reveal the phage-bacteria infection networks of the healthy human gut (Marbouty et al., Elife 2021). The student will also benefit from established collaboration with phage biology specialist Laurent Debarbieux at Pasteur Institute (ANR PhastGut). To ensure state of the art genetic and cellular microscopy analyses we will team up with the Cell cycle and chromosome dynamics (C3D) group headed by Olivier Espeli at College de France. In the recent years, RSG and C3D groups have successfully collaborated for the study of E. coli genome organization (Lioy, Cournac et al Cell 2018; Conin et al NAR 2022). Genetic design, cell biology observations, physiological / phenotypical consequences of Ndd production will be performed and analysed in the C3D group. Their expertise will be important to insure a comprehensive analysis of some of our expected results.

The genomic experiments planned during the PhD will require computational analysis. For these tasks, the student will be help by permanent bioinformatician researcher in the lab (Axel Cournac), a specialist in Hi-C analysis. He/She will also be able to apply for support from the HUB, a pool of engineer dedicated to support IP scientist over short-term period of time with computational assistance, if needed. But, more importantly, she will follow the IP courses of bioinformatics, biostatistics, Linux, biopython, that take place twice a year or so. Therefore, he/she will be able to handle and process her genomic data up to a certain point, facilitating their immediate interpretation.

Thèses actuellement en cours dans l'équipe

Tous les encadrements doivent être indiqués (y compris les co-directions avec un autre HDR pour des doctorants d'une autre ED, et les encadrements dans le cadre de programmes doctoraux tels qu'IPV, FDV...)

Nom et Prénom du doctorant	Directeur(s) de thèse	Année de 1ère inscription	ED	Financement
Meneu Léa	Koszul	2020	CdV	ERC
Girard Fabien	Koszul, Cournac	2020	CdV	ENS
Gradit	Cournac	2021	CdV	ANR JCJC Cournac
Bignaud	Marbouty	2020	CdV	ENS
Conti Devon	Marbouty, Debarbieux	2021	CdV	ANR PhastGut Debarbieux/Marbouty
Bruder Emma	Espeli	2020	BioSPC	MESRI

Trois publications récentes du directeur de thèse (du co-directeur ou du co-encadrant s'il y a lieu). Mettre en gras le nom du directeur de thèse.

1. Closed and High-Quality Bacterial Genome Sequences of the Oligo-Mouse-Microbiota Community. Lamy-Besnier Q, Koszul R, Debarbieux L, **Marbouty M**#. Microbiol Resour Announc. 2021 Apr 29;10(17):e01396-20. doi: 10.1128/MRA.01396-20. PMID: 33927045.
2. **Marbouty M**#, Thierry A, Millot GA, Koszul R#. MetaHiC phage-bacteria infection network reveals active cycling phages of the healthy human gut. *eLife* 2021;10:e60608. https://doi.org/10.7554/eLife.60608.
3. Böhm K, Giacomelli G, Schmidt A, Imhof A, Koszul R, **Marbouty M**&, Brankamp M&. Chromosome organization by a conserved condensin-ParB system in the actinobacterium Corynebacterium glutamicum. *Nat Commun* 2020;11:1485. https://doi.org/10.1038/s41467-020-15238-4.

1. Extended sister-chromosome catenation leads to massive reorganization of the E. coli genome. Conin B, Billault-Chaumartin I, El Sayyed H, Quenech'Du N, Cockram C, Koszul R, **Espéli O**. *Nucleic Acids Res*. 2022 Feb 25:gkac105. doi: 10.1093/nar/gkac105. Online ahead of print. PMID: 35212387
2. The Crohn's disease-related bacterial strain LF82 assembles biofilm-like communities to protect itself from phagolysosomal attack. Prudent V, Demarre G, Vazeille E, Wery M, Quenech'Du N, Ravet A, Dauverd-Girault J, van Dijk E, Bringer MA, Descrimes M, Barnich N, Rimsky S, Morillon A, **Espéli O**. *Commun Biol*. 2021 May 25;4(1):627. doi: 10.1038/s42003-021-02161-7. PMID: 34035436
3. Intracellular Positioning Systems Limit the Entropic Eviction of Secondary Replicons Toward the Nucleoid Edges in Bacterial Cells. Planchenault C, Pons MC, Schiavon C, Siguier P, Rech J, Guynet C, Dauverd-Girault J, Cury J, Rocha EPC, Junier I, Cornet F, **Espéli O**. *J Mol Biol.* 2020 Feb 7;432(3):745-761. doi: 10.1016/j.jmb.2019.11.027. Epub 2020 Jan 11. PMID: 31931015

Docteurs encadrés par le directeur de thèse ayant soutenu entre la date de dépôt de ce dossier et il y a 5 ans et publications relatives à leur sujet de thèse. Mettre en gras le nom du directeur de thèse et celui du docteur.

Sous la direction de Martial Marbouty

Nom Prénom :**Foutel-Rodier Théo**

Date de soutenance : 10/09/21

Durée de thèse (en mois) : 48 mois

Ecole Doctorale : CdV

Publications :

MetaTOR: A Computational Pipeline to Recover High-Quality Metagenomic Bins from Mammalian Gut Proximity-Ligation (meta3C) Libraries. Baudry L**, Foutel-Rodier T**, Thierry A, Koszul R#, **Marbouty M**#. Front Genet. 2019 Aug 20;10:753. doi: 10.3389/fgene.2019.00753. eCollection 2019. PMID: 31481973.

Generation of a Metagenomics Proximity Ligation 3C Library of a Mammalian Gut Microbiota. **Foutel-Rodier T**, Thierry A, Koszul R#, **Marbouty M**#. Methods Enzymol. 2018;612:183-195. doi: 10.1016/bs.mie.2018.08.001. Epub 2018 Sep 18. PMID: 30502941.

Nom Prénom : **Lamy-Besnier Quentin**

Date de soutenance : 03/12/21
Durée de thèse (en mois) : 39 mois
Ecole Doctorale :CdV

Publications :

Closed and High-Quality Bacterial Genome Sequences of the Oligo-Mouse-Microbiota Community. **Lamy-Besnier Q**, Koszul R, Debarbieux L, **Marbouty M**#. Microbiol Resour Announc. 2021 Apr 29;10(17):e01396-20. doi: 10.1128/MRA.01396-20. PMID: 33927045.

Sous la direction d'Olivier Espéli

Nom Prénom : **Brenna Conin**

Date de soutenance : 12/12/21
Durée de thèse (en mois) : 39 mois
Ecole Doctorale :CdV (co direction avec Romain Koszul)

Publication :
Extended sister-chromosome catenation leads to massive reorganization of the E. coli genome. **Conin B**, Billault-Chaumartin I, El Sayyed H, Quenech'Du N, Cockram C, Koszul R, **Espéli O**. Nucleic Acids Res. 2022 Feb 25:gkac105. doi: 10.1093/nar/gkac105. Online ahead of print. PMID: 35212387

Nom Prénom : **Victoria Prudent**

Date de soutenance : 13/12/29
Durée de thèse (en mois) : 48 mois
Ecole Doctorale :BioSPC

Publications :
The Crohn's disease-related bacterial strain LF82 assembles biofilm-like communities to protect itself from phagolysosomal attack.

Prudent V, Demarre G, Vazeille E, Wery M, Quenech'Du N, Ravet A, Dauverd-Girault J, van Dijk E, Bringer MA, Descrimes M, Barnich N, Rimsky S, Morillon A, **Espéli O**. Commun Biol. 2021 May 25;4(1):627. doi: 10.1038/s42003-021-02161-7.

The Crohn's disease-associated Escherichia coli strain LF82 relies on SOS and stringent responses to survive, multiply and tolerate antibiotics within macrophages.

Demarre G*, **Prudent V***, Schenk H, Rousseau E, Bringer MA, Barnich N, Tran Van Nhieu G, Rimsky S, De Monte S**, Espéli O**. PLoS Pathog. 2019 Nov 14;15(11):e1008123. doi: 10.1371/journal.ppat.1008123. eCollection 2019 Nov. (* co first author)

Imaging the Cell Cycle of Pathogen E. coli During Growth in Macrophage.
Demarre G, **Prudent V, Espéli O**. Methods Mol Biol. 2017;1624:227-236. doi: 10.1007/978-1-4939-7098-8_17.

Nom Prénom : **Elise Vickridge**

Date de soutenance : 22/06/18
Durée de thèse (en mois) : 44 mois
Ecole Doctorale :SDSV

Publications :
Revealing Sister Chromatid Interactions with the loxP/Cre Recombination Assay.
Vickridge E, Planchenault C, **Espéli O**. Methods Mol Biol. 2017;1624:29-37. doi: 10.1007/978-1-4939-7098-8_3.

Management of E. coli sister chromatid cohesion in response to genotoxic stress.

Vickridge E, Planchenault C, Cockram C, Junceda IG, **Espéli O**. Nat Commun. 2017 Mar 6;8:14618. doi: 10.1038/ncomms14618.

Mapping Topoisomerase IV Binding and Activity Sites on the E. coli Genome.
El Sayyed H, Le Chat L, Lebailly E, **Vickridge E**, Pages C, Cornet F, Cosentino Lagomarsino M, **Espéli O**. PLoS Genet. 2016 May 12;12(5):e1006025. doi: 10.1371/journal.pgen.1006025. eCollection 2016 May. PMID: 27171414