



MASTER 2 BMC PARCOURS GENOPATH ANNÉE 2023-2024

Titre du sujet de stage: RNA chaperons involvement in the regulation of *Dickeya dadantii* virulence

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Sujet de stage :

Small RNAs have been identified as central players reprogramming gene expression where they allow environmental adaptation and fitness. In bacteria, this post-transcriptional regulation classically occurs through base-pairing interactions between regulatory RNAs (sRNAs) and mRNAs, which are often assisted by specialized RNA-binding proteins called RNA chaperones such as Hfq or ProQ. These post-transcriptional regulations have been widely involved in the control of virulence factors in various bacteria (Djapgne et al, 2021 doi: 10.3389/fcimb.2021.604511). In the context of plant pathogenic bacteria where there is no method to treat diseases because antibiotics cannot be used, the development of alternative solutions is necessary. Interestingly, it has recently been suggested that regulatory RNAs can be used as tools for bioremediation and biomonitoring to achieve sustained eco-agriculture (Barathi et al https://doi.org/10.3390/ijms24021041).

Our team focuses its research on *Dickeya dadantii* which cause soft rot symptoms on a broad range of angiosperm plants including potato (Mansfield et al., 2012, doi: 10.1111/j.1364-3703.2012.00804.x). We have previously shown that the loss of Hfq or ProQ resulted in a drastic reduction in virulence. This phenotype was associated with alterations in several virulence determinants including cellulase, protease and pectate lyase production, motility, and adhesion, suggesting that Hfq and ProQ should directly or indirectly regulate these functions post-transcriptionally using sRNAs (Leonard, et al 2021, doi: 10.3389/fmicb.2021.687484. eCollection 2021). In parallel, our team identified the small transcript

repertoire of *D. dadantii* (Leonard, et al 2019, doi: 10.1093/nar/gkz485). The aim of this internship is to understand mechanisms by which Hfq and ProQ control virulence factors.

First, you will perform RNA-seq analyses of the *hfq* mutant, *proQ* mutant and wild-type strain in order to determine which genes have their expression level altered by the loss of Hfq, by the loss of ProQ (mRNAs and sRNAs). Then, you will select candidates and validate them using RT-qPCR and phenotypic analyses. Finally, you will start the characterization of the mechanism of action of Hfq and ProQ action by answering the following questions: which RNAs interact directly with Hfq, with ProQ? Which sRNA regulates mRNAs encoding virulence factors?

Technologies utilisées: molecular biology (RNA extraction, RT-qPCR, PCR, RNA-protein-immunoprecipitation analysis, genetics (reverse genetics), data analysis (bioinformatic tools), microbiology (culture, phenotypic tests, virulence test).

Mots clés: Post-transcriptional regulation, RNA-seq, non coding RNAs, RNA chaperone

Publications d'intérêt :

Djapgne et al, 2021 doi: 10.3389/fcimb.2021.604511

Mansfield et al., 2012, doi: 10.1111/j.1364-3703.2012.00804.x

Leonard, et al 2021, doi: 10.3389/fmicb.2021.687484. eCollection 2021

Leonard, et al 2019, doi: 10.1093/nar/gkz485 Barathi et al https://doi.org/10.3390/ijms24021041