M2 Internship / 2022

Regulation of bacterial toxin-antitoxin systems

Context

Our team is interested in type I toxin-antitoxin systems (TAT1) from pathogenic bacteria. In these systems, the antitoxin is an antisense RNA that inhibits of translation and/or promotes degradation of the toxin mRNA. In the absence of the antitoxin, the toxin interferes with the bacterial membrane resulting in the death of the cell producing it. To understand the precise mechanisms of regulation of these systems, we use a novel technique, the Fastbac-seq method (Functional AnalysiS of Toxin-Antitoxin Systems in BACteria by Deep Sequencing)[1] that combines random mutagenesis with next generation sequencing. This method allows to ascertain the toxicity of a peptide and permits the identification of mutations that inhibit the expression or function of the toxin. The broad distribution of these systems among pathogenic bacteria together with their ability to cause bacterial death makes TAT1 great candidates for the development of new antibiotics. Understanding their regulation and mechanism of action is crucial for exploiting them as a new source of antimicrobials.

[1]Masachis S, Tourasse NJ, Lays C, Faucher M, Chabas S, Iost I, Darfeuille F. *Elife*. **2019** Aug 14;8. pii: e47549. doi: 10.7554/eLife.47549.

Objectives

Validate inactivating mutants identified by FastBac-seq in selected T1TAs

Investigate the mechanism by which these mutants inactivate toxicity

Study the expression of selected systems

Methodology

Genetic constructs in *Escherichia coli*

DNA transformation and recombination on the bacterial chromosome

Molecular biology techniques (*eg*. Northern blot, Western blot)

Bacteriology techniques (*eg*. growth and viability assays)

Prerequisite

Molecular biology; Bacteriology

Key words

Molecular microbiology; Post-transcriptional regulation; regulatory RNAs

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