SUJET DE THESE / PhD SUBJECT

OFFRE D'ALLOCATION DE THESE / PhD GRANT

ÉCOLE DOCTORALE SCIENCES EXACTES ET LEURS APPLICATIONS - ED 211 / NATURAL SCIENCES DOCTORAL SCHOOL

Avenue de l'université BP 1155 64 013 PAU Cedex – France

TITRE / TITLE:

Genetic determinisms involved in mercury methylation by sulfate reducing bacteria

ABSTRACT:

Mercury (Hg) is a persistent pollutant in the environment, highly volatile and able to be converted into highly toxic methylmercury (MeHg). MeHg is a serious threat as it is a neurotoxic compound, which is bioaccumulated and bioamplified in food webs. Microorganisms play a central role in MeHg conversion, either directly by controlling Hg methylation and MeHg degradation. Although *hgcA* and *hgcA* genes have been identified as necessary for Hg methylation, today, the methylation process cannot be fully explained. Together with environmental factors, other genetic determinisms are suspected to be involved in mercury methylation. To date, little is known about the cellular and environmental mechanisms favouring MeHg production, and Hg methylation processes are far from being deciphered.

Our team at IPREM lab aims to characterize Hg methylation at cellular level, from Hg recognition by the cell to Hg export, including methylation steps. We intent to decipher the role of Hg cell trafficking in the Hg methylation process in two Sulfate Reducing Bacteria (SRB) model strains, *Desulfovibrio hydrargyri* BerOc1, able to methylate Hg and demethylate MeHg and *Desulfovibrio alaskensis* G20, able to exclusively demethylate MeHg. The objective of the thesis is to generate *D. hydrargyri* BercOc1 mutants deleted in genes involved in either methylation, sensing, and export, and their heterologous expression in G20. The effect in methylation and demethylation will be then evaluated on mutants and compared to wild type strains.

Keywords: mercury methylation, sulfate-reducing bacteria, genetics of SRB

CONDITIONS D'EXERCICE / WORKING CONDITIONS

Laboratoire : : Institut des Sciences Analytiques et de Physico-chimie pour l'Environnement et les Matériaux (IPREM UMR 5254, Pau)

Site web : ***https://iprem.univ-pau.fr/fr/index.html***

Directeur de thèse (PhD Supervisor): Marisol Goñi Urriza

The proposed PhD is part of the project 'GO-BEAM' (Go inside a bacterial cell methylating Mercury) funded by E2S-UPPA from 2018 to 2021. GO-BEAM, selected as a 'Key Scientific Challenges E2S-UPPA' (http://e2s-uppa.eu/en/index.html) is a collaborative and transdisciplinary project involving genetic microbiology, analytical chemistry, imaging and spectroscopy. The objective of the project is to improve the understanding of the Hg methylation/demethylayion processes at the cell level. 2 PhD and 1 Post-Doctorate are funded for the GO-BEAM project: PhD1 on analytical and imaging, PhD2 on genetic microbiology and physiological studies (the present proposition) and the Post-Doc on both imaging and spectroscopy. PhD1, PhD2 and Post-Doc will all start in 2018.

Scientific team: MP Isaure, M Goni, M Monperrus, B Khalfaoui-Hassani, R Guyoneaud, C. Gassie, 2 PhD students, 1 post-doc.

Lieu (Place) : IPREM, Pau

Date début (start): October 2018	**Durée (duration):** 3 years

Employeur (employer): Université de Pau et des Pays de l'Adour (UPPA)

Salaire mensuel brut (monthly salary before taxes): 1868 € (*doctoral contract UPPA, according to E2S Key scientific challenges project, including 96h of teaching during the three years)*

SAVOIR-FAIRE DU LABORATOIRE / HOST LABORATORY PROFILE

Analytical chemistry, mass spectrometry, X-ray absorption spectroscopy, Imaging, Microbiology, Physiology, Genetics, Microbial Ecology, Environmental Microbiology

MISSION - ACTIVITES PRINCIPALES / MISSION – PRINCIPAL ACTIVITIES

I. *Scientific Context*

Mercury (Hg) is one of the major contaminants at the global scale. It is persistent, highly volatile and is able to convert into highly toxic methylmercury (CH_3Hg or MeHg), a strong neurotoxic. The production of highly toxic methylmercury (MeHg) is mediated by microorganisms but little is known about the cellular and environmental mechanisms favoring MeHg production. Understanding the biotransformation processes of Hg by microorganisms is thus a key for Hg risk assessment in ecosystems and human health. A few years ago, Parks et *al.* (2013) identified two genes, *hgcA* and *hgcB* required for Hg methylation. However, strains carrying *hgcAB* genes produce methylmercury at different rates, partly depending on their physiological state and environmental parameters (Goñi-Urriza *et al.* 2015). Furthermore, our results coupled with other studies with different mercury concentrations suggest that mercury methylation can be regulated by the export and/or by the intracellular contents of mercury.

II. *Objectives*

The main objective of this PhD thesis is to evaluate the role of various genes (involved in Hg recognition, methylation and export) in Hg methylation/demethylation.

III. *Work plan*

The thesis work will be devoted at characterizing the role of candidate genes in Hg methylation. It will be assessed through gene deletion and complementation in *D. hydrargyri* BerOc1 and through heterologous expression in the non methylating strain G20 as follows:

- **Gene deletion**: Specific gene knockout in *D. hydrargyri* BerOc1 will be performed using one-step homologous double-crossover procedure. Genetic tools have been already optimized for *D. hydrargyri* BerOc1.
- **Over-expression of genes of interest**: The genes encoding *hgcAB* or the export systems will be overexpressed in wild type *D. hydrargyri* BerOc1 in order to evaluate the limits of mercury methylation and the effects on MeHg demethylation.
- **Heterologous expression** of genes of interest in G20: The genes encoding *hgcAB* or the export systems will be co-expressed in the non-methylating strain *D. alakansis* G20.

He/She will also perform physiological studies to understand the changes in growing, gene expression and mercury methylation and speciation. The gene deletion and over-expression in BerOc1 as well as the heterologous expression in G20 will be done in close collaboration with Alain Dolla and Nathalie Pradel from MIO laboratory in Marseille.

The student will work with another PhD student involved in analytical chemistry and a post-doc, involved in X-ray imaging and X-ray Absorption Spectroscopies techniques. By applying isotopic tracer approaches and X-ray imaging on the wild type and mutants, the Hg methylation potentials and localization will be characterized in order to decipher the role of the studied genes on Hg pathways.

The phD student will also participate to teaching activities at the undergraduate level (96h/3 years).

IV. *Literature References*

Goñi-Urriza M, Corsellis Y, Lanceleur L, Tessier E, Gury J, Monperrus M, Guyoneaud R. 2015. Relationships between bacterial energetic metabolism, mercury methylation potential and hgcA / hgcB gene expression in Desulfovibrio dechloroacetivorans BerOc1. *Environ Sci Pol Res* 22, 13764.

Parks JM, Johs A, Podar M, Bridou R, Hurt Jr RA, Smith SD, Tomanicek SJ, Qian Y, Brown SD, Brandt CC, et al. 2013. The genetic basis for bacterial mercury methylation. Science 339: 1332-1335.

COMPETENCES REQUISES / REQUIRED COMPETENCES

Skills in microbial genetic, molecular biology and microbial physiology are required
The candidate should have a strong predilection for laboratory work.
The ideal candidate has a master degree in molecular biology, genetics or microbiology. He/She is rigorous and highly motivated. He/she must have a good English level and the capacity to work autonomously.
French spoken will be a plus (teaching activities)

CRITÈRES D'ÉVALUATION DE LA CANDIDATURE / CRITERIA USED TO SELECT CANDIDATE

Two steps selection process:
1st step:
- Evaluation of the applicants' cv
- Selected candidates will be contacted by mail before the **24/08**

2nd step: **28-29/08**
- Selected candidates will have 5 min to present their CV, 5 min to present their Master2 thesis and 5 min to present a comprehensive scientific view on the phD project
- This presentation will be followed by questions and discussion.

Criteria used in selection of the candidate:
- The candidate's motivation, scientific maturity and curiosity.
- Candidate's knowledge.
- Candidate's marks and rankings in Licence/undergraduate, M1 and M2.
- English proficiency
- Candidate's ability to present his work
- Professional experience of internship (s) in laboratory or other; any research work already carried out (reports, publications)

CONSTITUTION DU DOSSIER DE CANDIDATURE, DATE LIMITE DE DEPOT / REQUIRED DOSSIER, DATE

Application should be send by e-mail. The application should contain:
- CV
- Cover letter detailing candidate's motivations
- Candidate's Licence and MSc marks and ranking
- Reference letters
- Contact details (for 2 referees)

DATE LIMITE DE DEPOT DU DOSSIER (deadline):	**15/08/2018**

CONTACTS

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