Proposal for the 2020 Imagine International PhD program

Laboratory: Genetics of Mitochondrial Disorders
Head of laboratory: Agnès RÖTIG
Project and student supervisor: Agnès RÖTIG DR1
Number of HDR in the lab: 5
Field of research: Mitochondrial disorders, Iron homeostasis
Number and names of PhD students currently in the lab in 2020: 1st y, 2nd Y, 3rd y, 4th Y
Floriane PETIT, 3rd y managed by Agnès RÖTIG
Cérane CAFOURNET, 2nd y managed by Benedetta RUZZENENTE
Kaliopi CHATZOVOULOU, 2nd y managed by Julie STEFFANN
Number and names of PhD students under the Imagine program: 0
Project Title: Iron homeostasis in Friedreich ataxia and Neurodegeneration with Brain Iron Accumulation

Project description (Max 2 pages including abstract and publications)

ABSTRACT

Friedreich ataxia (FRDA) and Neurodegeneration with Brain Iron Accumulation (NBIA) are rare neurodegenerative diseases due to mutations in different genes but sharing a same mechanism of iron accumulation ascribed to abnormal recycling of transferrin (Tf), impaired transferrin receptor (TfR1) trafficking and a reduction of TfR1 palmitoylation related to decreased Coenzyme A (CoA) level. FRDA and several NBIA subtypes also result from various mitochondrial dysfunctions. This PhD thesis project aims to delineate the connection(s) between mitochondrial defect, iron homeostasis, CoA synthesis and post-translational protein modification by pamitoylation in FRDA and NBIA. The different work packages of this project consists to:

- investigate in patients' fibroblasts a poorly known mechanism of non-Tf bound iron (NTBI) uptake, the associated factors involved in and their anomalies,
- the mechanism that leads to impaired CoA homeostasis in FRDA and NBIA,
- better understand the shared cellular pathway(s) that are impaired in these diseases for identifying therapeutic targets and molecules.

PhD PROJECT

Friedreich ataxia (FRDA) result from a defect of the mitochondrial protein, frataxin, involved in the first steps of iron-sulfur cluster (ISC) synthesis. NBIA is genetically heterogeneous condition caused by mutations in 15 different genes involved in various cellular processes such as CoA biosynthesis and lipid metabolism, endosome recycling, autophagy and lysosomal activity. Despite genetic heterogeneity, these diseases share brain iron accumulation and cellular iron dysregulation. However, the mechanism of iron accumulation is not completely elucidated. We have reported that NBIA fibroblasts are unable to limit iron uptake that we ascribed to abnormal post-translational modification of TfR1 by palmitoylation (Drecourt et al, 2018). Surprisingly, FRDA fibroblasts present a very similar phenotype of iron dysregulation that we only partially described. The PhD project will aim to better understand this deregulation of iron homeostasis by using fibroblast of a series of patients with FRDA and NBIA.

Non-transferrin bound iron (NTBI) uptake in FRDA and NBIA

Cells acquire iron as transferrin (Tf)-bound (TBI) or non-Tf-bound iron (NTBI). We characterized the TBI uptake deregulation in FRDA and NBIA. Preliminary data showed that NTBI uptake is also deregulated in FRDA but almost nothing of NTBI uptake regulation is known. Moreover, we showed that NTBI uptake depends on palmitoylation of yet unknown proteins. The TBI and NTBI uptake will be characterized in FRDA and NBIA fibroblasts, the known NTBI transporters will be studied and their regulation will be investigated by protein biochemistry, transcriptomic (RNAseq) and proteomic analysis.

CoA homeostasis in FRDA and NBIA

Impaired CoA homeostasis and defective lipid metabolism seem to have a central role in NBIA and FRDA. Indeed, we demonstrated that frataxin defect indirectly result in decreased CoA synthesis. Nevertheless, only few NBIA genes are directly involved in CoA synthesis. The CoA content will be quantified in fibroblasts of NBIA related to various gene mutations. The effect of CoA on TBI and NTBI iron uptake and on iron content will be studied with the aim to identify the link between the NBIA genes and CoA content.

Therapeutic approach for FRDA and NBIA

We showed that artesunate, a derivative of artemisinin, enhanced TfR1 palmitoylation in fibroblasts from NBIA and FRDA patients. Moreover, artesunate lowered the iron content of FRDA fibroblasts. The next step is to determine if artesunate also lower iron content in NBIA fibroblasts. Moreover, molecules able to increase CoA content and/or protein palmitoylation will also be studied. We have already identified few of such molecules that represent interesting therapeutic molecules after repurposing. It should be mentioned that a large series of patients with FRDA and NBIA are followed in Necker hospital for future clinical trials.

The expected consequences of this proposal are twofold:
- it will contribute to better understand the deregulation of iron homeostasis in FRDA and NBIA, to identify shared cellular pathway(s) that are impaired in these diseases and to investigate yet poorly known alternative iron uptake,
- it will identify therapeutic targets and molecules for these diseases. Importantly, the expected results of this proposal will have potential not only for FRDA and NBIA but also for more frequent conditions such as frontotemporal dementia (FTD), Parkinson's disease (PD) and Alzheimer's disease (AD) also characterized by brain iron overload.

Lab members

Direction

Agnès RÖTIG	DR1 INSERM

Mitochondrial RNA and protein maturation

Metodi METODIEV	CRN INSERM
Benedetta RUZZENENTE	Senior Post-doc
Martin HORAK	Post-doc
Cérane CAFOURNET	PhD student
Lucas BIANCHI	IE CDD INSERM

Mitochondrial diseases and interferon response

Manuel SCHIFF	PU-PH
Alessandra PENNISI	PhD student)
Chloé FORAY	Engineer

Metabolic flows, gene therapy of metabolic diseases

Chris OTTOLENGHI	PU-PH
Clément PONTOIZEAU	AHU
Irina ROTARU	M2

Mitochondrial DNA replication during embryo-fetal development

Jean-Paul BONNEFONT	PU-PH
Julie STEFFANN	PU-PH
Kaliopi CHATZOVOULOU	PhD student

Iron homeostasis in Friedreich ataxia and NBIA

Mirella LO SCRUTADO	Post-doc
Floriane PETIT	PhD student
Chloé ANGELINI	M2 student

Reference Center for Mitochondrial Disorders

Jean-Paul BONNEFONT	PU-PH

Major Publications

Pulman et al. Mutations in the MRPS28 gene encoding the small mitoribosomal subunit protein bS1m in a patient with intrauterine growth retardation, craniofacial dysmorphism and multisystemic involvement. Hum Mol Genet. 2019 28:1445-1462 **doi: 10.1093/hmg/ddy441**

Drecourt A et al. Impaired Transferrin Receptor Palmitoylation and Recycling in Neurodegeneration with Brain Iron Accumulation. Am J Hum Genet 2018 102:266–277. **doi: 10.1016/j.ajhg.2018.01.003**

Dhir A et al. Mitochondrial double-stranded RNA triggers antiviral signalling in humans. Nature. 2018 560:238-242 **doi: 10.1038/s41586-018-0363-0**

Gardeitchik T **et al**. Bi-allelic Mutations in the Mitochondrial Ribosomal Protein MRPS2 Cause Sensorineural Hearing Loss, Hypoglycemia, and Multiple OXPHOS Complex Deficiencies. Am J Hum Genet. 2018 102:685-69. **doi: 10.1016/j.ajhg.2018.02.012**

Lake et al. Biallelic Mutations in MRPS34 Lead to Instability of the Small Mitoribosomal Subunit and Leigh Syndrome. Am J Hum Genet. 2018 102:713. **doi: 10.1016/j.ajhg.2018.03.015**

Metodiev et al. Recessive Mutations in TRMT10C Cause Defects in Mitochondrial RNA Processing and Multiple Respiratory Chain Deficiencies. Am J Hum Genet. 2016 98:993-1000. **doi: 10.1016/j.ajhg.2016.06.013**

Sánchez-Caballero L et al. Mutations in Complex I Assembly Factor TMEM126B Result in Muscle Weakness and Isolated Complex I Deficiency. Am J Hum Genet. 2016, 99:208-16. **doi: 10.1016/j.ajhg.2016.05.022**