M1-Molecular and Cellular Biology (MCB) Internship Proposal Form Chemistry-Biology Department

Laboratory Address and Affiliation:

Institut de Biologie Structurale (IBS) Univ. Grenoble Alpes, CEA, CNRS 71 avenue des Martyrs CS 10090 38044 Grenoble Cedex 9 France www.ibs.fr

Laboratory/Team Research area (Keyword)

IBS/ Groupe Membrane and pathogens https://www.ibs.fr/research/research-groups/membrane-and-pathogens-group-f-fieschi/ Theme: NADPH Oxidase

Summary of the Proposed Internship Project (10 lines)

Title:

Structural and functional characterization of recombinant human NADPH oxidase NOX2.

DESCRIPTION:

Found in phagocytic cells, NOX2/p22^{phox} assembly, a heterodimer of about 100 kDa size are mutually stabilizing while in the membrane. This membranous complex is a crucial system in the innate immune defense. NOX2 transfers electrons from NADPH to O₂ to produce superoxide anion *via* two hemes and a flavin cofactor. Activation of NOX2/p22^{phox} requires assembly with phosphorylated cytosolic factors (p47^{phox}/p67^{phox}/p40^{phox}) and the RacGDP/RacGDI complex. Such a complex NOX2 activation process, requiring so many factors to play their roles, is still mechanistically unclear at the molecular level. In the M1 internship, the student will benefit from a recently developed expression system of a recombinant version of the human NOX2/p22^{phox} complex. The purification of NOX2/p22^{phox} complex and biochemical characterization will be executed (activity test and assembly with cytosolic factors) and first quality analysis by negative staining electron microscopy. The goal at the level of the M1 is to set up the production of a sample compatible with further structural analysis by Cryo-Electron Microscopy.

Methodologies and/or Techniques to be used

Cell culture, Membrane protein purification, Activity test (redox biochemistry), Insertion of membrane protein into nanodisc, preparation of Electron microscopy Grid, potentially interaction analysis using SPR (surface plasmon resonance).

Person to contact:

Name: Isabelle Petit-Hartlein & Franck Fieschi Phone: 04 57 42 85 92 E-mail: isabelle.petit-hartlein@ibs.fr

Additional information

When you apply, please send your CV and your marks for the L2 and L3 years of your study.