

Internship project Master 2 Year 2017-2018

Laboratory/Institute: Institut de Biologie Structurale
Team: Microscopie Electronique et Méthodes

Director: Pr. Winfried Weissenhorn
Head of the team: Dr. Guy Schoehn

Name and status of the scientist in charge of the project: Hélène Malet

Address: IBS, 71, avenue des Martyrs, CS 10090 38044 Grenoble

Phone: 04. 57. 42. 87. 77

e-mail: helene.malet@ibs.fr

Title of the project: Structural characterization of Hantaan virus nucleocapsid

Objectives (up to 3 lines):

Hantaviruses are life-threatening viruses closely related to Influenza virus. The aim of the project will be to analyse by cryo-electron microscopy the structure of their nucleocapsids, which are essential complexes implicated in packaging and protection of the viral genome. As nucleocapsids are essential parts of Hantavirus replication and transcription machineries, determining this structure will considerably improve our knowledge on these key steps of the viral cycle.

Abstract (up to 10 lines):

Hantaviruses belong to the Bunyaviridae family, a large family of viruses that possess a segmented RNA genome of negative sense polarity. Each genome segment is enwrapped with the viral nucleoprotein N forming a ribonucleoprotein complex, called the nucleocapsid. This complex protects the viral genetic information from the environment while providing a flexible helical template for viral transcription and replication by the viral RNA polymerase. As a unique structure in nucleic acid biology, the nucleocapsid constitutes an attractive potential target for antiviral drugs without harmful side effects. The proposed Master II project will consist in the structural analysis Hantaan virus nucleocapsids. The intern will express recombinant Hantaan nucleocapsid in insect cells and optimize its purification in order to obtain an homogeneous complex suitable for cryo-electron microscopy analysis. We already have preliminary results showing that recombinant Hantaan nucleocapsid is a straight helix amenable to high-resolution by cryo-EM.

Methods (up to 3 lines):

Following expression in insect cells and purification, the intern will collect negative stain and cryo electron microscopy data and perform EM image processing to determine the 3D structure of Hantaan nucleocapsid. He will benefit from the EM expertise in the group and will have access and training to the IBS state-of-the art EM facility, notably an FEI Polara microscope equipped with a Gatan K2 direct electron detector.

Up to 3 relevant publications of the team:

- . Gutsche I, Desfosses A, Effantin G, Ling WL, Haupt M, Ruigrok RW, Sachse C, **Schoehn G**. Structural virology. Near-atomic cryo-EM structure of the helical measles virus nucleocapsid. **Science** 348, 704–707 (2015). 1
- . Gerlach, P*, **Malet, H***, Cusack, S & Reguera, J Structural Insights into Bunyavirus Replication and Its Regulation by the vRNA Promoter. **Cell** 161, 1267–1279 (2015). *Co-first author 2
- . Reguera J, **Malet H**, Weber F, Cusack S. Structural basis for encapsidation of genomic RNA by La Crosse Orthobunyavirus nucleoprotein. **Proc Natl Acad Sci U S A**. 2013 Apr 30;110:7246-51. 3

Requested domains of expertise (up to 5 keywords): Structural biology, electron microscopy, biochemistry, virus, nucleocapsid.