Funded PhD project in Integrated Structural & Cell Biology in Grenoble, France

Title of the PhD project:
Env-mAb: Structure of the HIV-1 envelope glycoprotein in complex with a novel bnAb

PhD supervisors:
Winfried Weissenhorn winfried.weissenhorn@ibs.fr & Grégory Effantin gregory.effantin@ibs.fr

Host laboratory:
Institut de Biologie Structurale, EBEV and MEM groups

Project summary:
The HIV-1 envelope (Env) glycoprotein catalyses virus entry by fusing the viral envelope with cellular membranes, thereby establishing infection. In order to merge viral and cellular membranes, Env undergoes a series of conformational changes triggered by its binding to the cellular receptors CD4 and CCR5/CXR4. Env is also the main target of broadly neutralizing antibodies (bnAbs) that must be induced by potential immunogens to provide protection via vaccination. In order to design the best possible Env immunogen reverse vaccinology approaches are applied which include understanding the structural basis of Env recognition by different classes of bnAbs, which will guide engineering Env for optimal antigen presentation. Here we propose to solve the structure of Env in complex with a novel bnAb that recognizes the interface between gp120 and gp41 by cryo-electron microscopy. Furthermore, we will focus on a second epitope within the gp41 membrane proximal external region (MPER) that is not present in current high-resolution structures of Env. We have designed a soluble version of Env trimers including MPER that form native closed trimers, which bind bnAbs with high affinity. Secondly, we propose to solve the structure of a stabilized consensus C clade Env. Thirdly, we propose to solve the structure of complete Env including MPER, the transmembrane region and the cytoplasmic domain to high resolution by cryo-EM, which will help to understand the structural basis of the membrane-anchored native Env conformation. Together, our structural approaches will provide important new insights into (i) Env recognition by a novel bnAb and (ii) into the structure of MPER within a native Env trimer, which will have important implications for vaccine development.

Required skills:
Protein biochemistry, structural biology and a strong interest in cryo-electron microscopy.

Student role:
The student will have the opportunity to obtain training in mammalian expression techniques. The student will practically lead the project from the purification step on. He/she will be trained in the purification of Env and in the purification of the antibodies and Fab production from antibodies. The student will purify the complexes by SEC for further electron microscopy analysis. The student will be trained on all aspects of electron microscopy. The student will learn to prepare EM grids to test the samples first by negative stain. The obtained images will also enable the candidate to perform image analysis and therefore to learn the technique. The student will then be trained as well in all the steps needed to do cryo-electron microscopy (grid preparation, data acquisition, image analysis and atomic model building and refinement). For electron microscopy, the student will benefit from the access to the instruments of the IBS EM platform (T12, F20 and Glacios) and of the ESRF (Titan krios).

Keywords:
HIV-1, Env, bnAb, gp41 MPER, cryo-electron microscopy

Relevant publications of the team:
- Caillat C, Guilligay D, Torralba J, Friedrich N, Nieva JL, Trkola A, Chipot C, Dehez F, Weissenhorn W, (2020) Structure of HIV-1 gp41 with its membrane anchors targeted by neutralizing antibodies. bioRxiv 2020.11.12.379396; doi: https://doi.org/10.1101/2020.11.12.379396.
- Caillat C, Guilligay D, Sulbaran G, Weissenhorn W. Neutralizing Antibodies Targeting HIV-1 gp41. Viruses. 2020 Oct 23;12(11):1210. doi: 10.3390/v12111210.
- Pinto,D., Fenwick, C., Caillat, C., Silacci, C., Guseva, S., Dehez, F., Chipot, C., Barbieri, S., Minola, A., Jarrossay, D., Tomaras, G.D., Shen, X., Riva, A., Tarkowski, M., Schwartz, O., Bruel, T., Dufloo, J., Seaman, M.S., Montefiori, D.C., Lanzavecchia, A., Corti, D., Pantaleo, G. and Weissenhorn, W. (2019) Structural Basis for Broad HIV-1 Neutralization by the MPER-Specific Human Broadly Neutralizing Antibody LN01. Cell Host & Microbe, 26(5):623-637.e8. doi: 10.1016/j.chom.2019.09.016.
- Effantin, G., Estrozi, L., Ashman, N., Renesto, P., Stanke, N., Lindemann, D., Schoehn, G., and Weissenhorn, W. (2016) Molecular architecture of the retrovirus prototype foamy virus. PLoS Pathog. 12(7):e1005721.