

Master 2 Internship

## Multiview reconstruction of particles with ninefold cylindrical symmetry in fluorescence imaging

Fluorescence microscopy has been revolutionized by the recent development of super-resolution techniques that overcame the resolution limits of conventional optical microscopes. It is now possible to image the protein structure of small fundamental cellular units such as macromolecular assemblies, which were not observable in live cells until recent years. This opens a new field of investigation that has been growing very rapidly [5, 6]. However, intrinsic physical and biological limitations still restrict the impact of these techniques: Firstly, the 3D resolution in fluorescence microscopy is strongly anisotropic (the axial resolution is 3 to 5 times lower than in the lateral plane), and secondly, the fluorescent proteins do not cover uniformly the structures of interest such that the observation of a particle is incomplete.

To overcome these limitations, this internship will take place in the context of a single particle reconstruction (SPR) project<sup>1</sup>. The principle of SPR is to acquire images that contain a large number of randomly oriented copies of a rigid particle to reconstruct a single model of the particle (Fig. 1). The combination of multiple views allows us to obtain high isotropic resolution, and to compensate for the partial labelling in the input data.

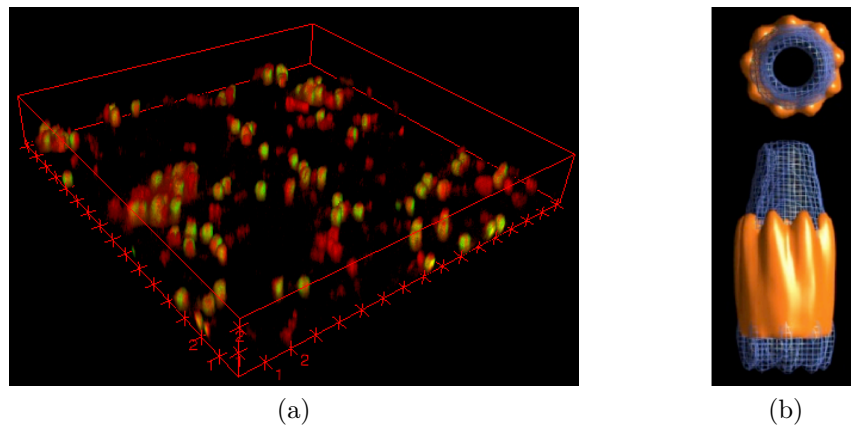


Figure 1: Principle of single particle reconstruction. (a): 3D image containing multiples copies of the same particle, called centriole; the axial resolution is low and the labelling of the particles is incomplete. (b): Reconstructions of a single model of the centriole with high isotropic resolution and homogeneous labelling.

If the orientations of the particles are known, SPR turns out to be a multiview reconstruction problem for which efficient solutions exist [1]. However, the particles are randomly rotated in the sample and we

<sup>1</sup>Funded by the French National Research Agency (ANR), through the "SP-Fluo" project

have no prior knowledge about their pose in the acquired images. The main challenge of SPR is then to estimate the orientations jointly with the reconstruction. We have developed methods in our group to address this problem [2]. However our current solution can be computationally expensive and requires the tuning of hyperparameters, depending on the type of imaging modality and of particle geometry. This can be a limitation for practical applications in biology.

Therefore, in this internship we will focus on a particular case to simplify the problem: we will work on a specific particle called centriole. It is a large multi-protein complex that is present in most eukaryotic cells, and it is essential for cilia, flagella and centrosomes formation. There is a large recent literature on the study of this macromolecular complex [3]. The centriole has a barrel shape with a ninefold cylindrically symmetric structure [4]. This type of geometry is common to several important macromolecular assemblies studied in structural biology. This knowledge about the shape of the particle can be translated in a powerful prior for the estimation of the orientations. Thus, the primary goal of the internship will be to develop a deep learning based method for orientation estimation of the centriole that leverages the ninefold cylindrical symmetry structure. Depending on the progress during the internship, the intern could also address the first step of the reconstruction pipeline, that is detection of the particles in the input images (Cf. Figure 1.a). A method based on 3D deep neural network has already been developed in a previous internship. We would like to investigate different strategies based 2D neural networks for this task, in order to improve the efficiency of the method.

This project will be realized in collaboration with biologists of the group of Prof. Paul Guichard in University of Geneva (<https://cellbio.unige.ch/research/paul-guichard/>). We have at our disposal large datasets of centrioles obtained with different fluorescence microscopy modalities that will be used to validate the performance of the methods.

## Working environment

The intern will be a member of the IMAGeS team (<http://images.icube.unistra.fr/>) in the ICube laboratory in Illkirch. The internship will begin between February and May 2022, for a period of 6 months.

Supervisors: Denis Fortun (CNRS researcher, ICube, [dfortun@unistra.fr](mailto:dfortun@unistra.fr)), Etienne Baudrier (Assistant Professor, University of Strasbourg, [baudrier@unistra.fr](mailto:baudrier@unistra.fr)).

## Profile of the candidate

- Second year of Master studies in one of the following fields: computer science, applied mathematics, machine learning
- Good programming skills (the coding language will be Python)
- An interest for biomedical applications is welcome

## References

- [1] D. Fortun, P. Guichard, N. Chu, and M. Unser. Reconstruction from multiple poses in fluorescence imaging: Proof of concept. *IEEE Journal of Selected Topics in Signal Processing*, 10(1):61–70, 2016.
- [2] D. Fortun, P. Guichard, V. Hamel, C. Sorzano, N. Banterle, P. Gonczy, and M. Unser. Reconstruction from multiple particles for 3D isotropic resolution in fluorescence microscopy. *IEEE Transactions on Medical Imaging*, 37(5):1235–1246, 2018.

- [3] M. Le Guennec, N. Klena, D. Gambarotto, M. H. Laporte, A.-M. Tassin, H. Van den Hoek, P. S. Erdmann, M. Schaffer, L. Kovacik, S. Borgers, et al. A helical inner scaffold provides a structural basis for centriole cohesion. *Science advances*, 6(7):eaaz4137, 2020.
- [4] M. LeGuennec, N. Klena, G. Aeschlimann, V. Hamel, and P. Guichard. Overview of the centriole architecture. *Current Opinion in Structural Biology*, 66:58–65, 2021.
- [5] D. Salas, A. Le Gall, J. Fiche, A. Valeri, Y. Ke, P. Bron, G. Bellot, and M. Nollmann. Angular reconstitution-based 3d reconstructions of nanomolecular structures from superresolution light-microscopy images. *Proceedings of the National Academy of Sciences*, 114(35):9273–9278, 2017.
- [6] C. Sieben, N. Banterle, K. Douglass, P. Gonczy, and S. Manley. Multicolor single particle reconstruction of protein complexes. *Nature methods*, 15:777–780, 2017.