4D Imaging Techniques in Liquid Transmission Microscopy

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Electron microscopy (EM) is a technique that exploits the interaction between electrons and matter to produce high resolution images down to atomic level. To avoid undesired scattering in the electron path, EM samples are conventionally imaged in solid state under vacuum condition. Recently, this limit has been overcome by the realization of Liquidphase electron microscopy (LP EM), a technique that enables the analysis of samples not withstanding the vacuum. Therefore, LP EM paired with a camera with a very high frame rate allows tracking the motion and the evolution in time of particle in liquids and its dynamic processes.

The motion of the particle together with the presence of liquid significantly affect the quality of the images. Therefore, in order to be able to derive useful information about samples, the images have to go through postprocessing algorithms. First, noise has to be reduced from the original image. However, the more the noise is reduced, the less the details in the image. Consequently, a sharpening algorithm is necessary in order to restore – or even increase - the high accuracy of details of the image. After all these steps, images result to be less noisy and richer in details than the original ones. At this point of the process, images are ready to be analysed.

The idea at the base of the project consists in taking advantage of the motion of the particles recorded in videos, to overcome one of the main limits of the single particle analysis (SPA). SPA is a technique that uses images of thousands of particles of the same nature, to reconstruct 3D models of the particle itself. Images showing different profiles of the particle are clustered according to a similarity criterion, and put together, but problems may arise when angular orientations have to be assigned. Therefore, in order to solve this problem an a priori model can be used to facilitate the orientation assignments. 2D images are compared to the projections of the a priori model set in input, and as result an angular displacement is assigned to each profile. Consequently, the goodness of the a priori model inevitability biases the final result of the reconstruction. In this work, this limit is overcome by merging single particle analysis techniques with the dynamics of particles. Different particle profiles will be registered not only in single frames, but also across the videos, offering a very wide range of orientations. The angular orientation of each profile will be derived by tracking the evolution in time of the motion of each particle, and a priori models will no longer be needed.

In this scenario, the postprocessing phase assumes more and more importance inside the general algorithm, since sharpening single images can lead to uncover very specific features, useful to track the direction of the rotations. Furthermore, this project eliminates any boundaries to a priori knowledge and can be applied to a very wide range of particles.