A FULL 3D CORRELATIVE IMAGING WORKFLOW: CHALLENGES IN COMPUTER VISION

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1. CONTEXT

Imaging of biological matter across resolution scales entails the challenge of preserving the direct and unambiguous correlation of subject features from the macroscopic to the microscopic level. In this talk we present in detail the computer vision tasks part of a correlative imaging platform developed on a synchrotron (Beamline B24, Diamond Light Source, UK). The imaging platform was developed specifically for imaging cells in 3D under cryogenic conditions by using X-rays and visible light. It proposes an integrated, user-friendly platform for 3D correlative imaging of cells in vitreous ice by using super-resolution structured illumination microscopy in conjunction with soft X-ray tomography [1].

2. RESULTS AND CHALLENGES

We present here the different steps of the workflow used in Fig.1, where the analysis was performed using ec-CLEM [2]. The accurate matching of structures in both modalities required an analysis workflow with several intermediate images where error accumulation needs to be quantitatively estimated because of the important difference of scale and the unknown aspect of structure under study. Intermediate images were used for the relocation of the different field of view, and the last step was the 3D refine registration. This approach was demonstrated by studying the process of reovirus release from intracellular vesicles during the early stages of infection and identifying intracellular virus-induced structures [1]. The accuracy was found to be between 50 and 130 nanometers when using 15 points, and between 80 and 350 nanometers when using 5 points for 95% of the 3D field of view. Size of dataset to be processed and automation of number of steps are challenges for the workflow.

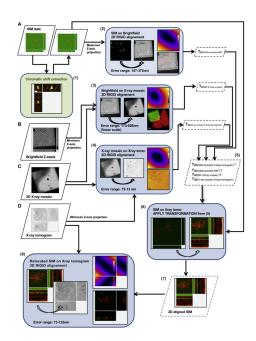


Fig. 1. *In silico* correlative workflow for 3D volume alignment of CryoSIM data on CryoSXT volumes. Figure from [1], under the Creative Commons CC-BY license.

3. REFERENCES

- [1] I. Kounatidis, M.L. Stanifer, M.A. Phillips, P. Paul-Gilloteaux, X. Heiligenstein, H. Wang, C.A. Okolo, T.M. Fish, M.C. Spink, D.I. Stuartand I. Davis, S. Boulant, J. M. Grimes, I. M. Dobbie, and M. Harkiolaki, "3d correlative cryo-structured illumination fluorescence and soft x-ray microscopy elucidates reovirus intracellular release pathway," *Cell*, vol. 182, pp. 515–530, July 2020.
- [2] P. Paul-Gilloteaux, X. Heiligenstein, M. Belle, M.C. Domart, B. Larijani, L. Collinson, G. Raposo, and J. Salamero, "ec-clem: flexible multidimensional registration software for correlative microscopies," *Nature Methods*, vol. 14, pp. 102–103, January 2017.

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