PoCA: a powerful visualization and quantification software for 3D single-molecule localization microscopy data

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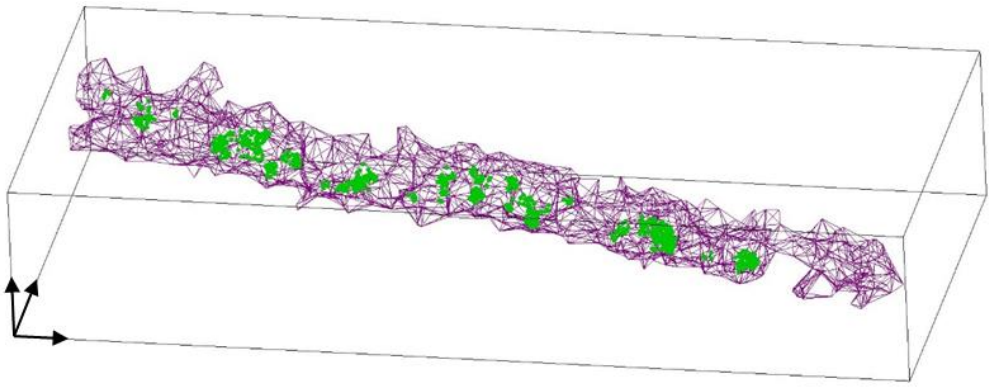

Figure 1: Multi-level segmentation of a mitochondria's shape and clusters performed in PoCA.

Over the last decade, single-molecule localization microscopy (SMLM) has revolutionized cell biology, making it possible to monitor molecular organization, dynamics and potential interactions with spatial resolution of a few nanometers. Based on pointillist image reconstructed from localizing single fluorescent emitters, SMLM represents a paradigm shift from conventional microscopy as it produces point clouds data instead of images. Nowadays, 3D SMLM data composed of millions of localizations are routinely acquired by biology research labs. However, despite recent efforts, there is still a critical challenge to provide a dedicated quantification tool allowing non-expert handling such a huge amount of data in 3D.

We here present PoCA (**Po**int **C**loud **A**nalyst), a powerful software for the visualization, manipulation and quantification of multidimensional SMLM data. Easily extensible through a plugin mechanism, PoCA integrates several state of the art quantitative analysis methods (ie. Tesselation, DBSCAN, K-Ripley's function etc…). It uses modern OpenGL libraries, offering several benefits: (i) the efficient interactive rendering of millions of 3D points; (ii) the integration of advanced rendering techniques, such as ambient occlusion, which strongly enhances user perception in crowded regions; (iii) the interactive object manipulation using mouse. Developed in C++, PoCA is thoroughly optimized, allowing to generate Voronoi diagrams of millions of 3D coordinates in a few minutes on a conventional computer [1, 2]. Finally, PoCA can execute Python scripts, facilitating prototyping and diffusion of new methods by and for the community.

[1] F. Levet et al, *SR-Tesseler: a method to segment and quantify localization-based superresolution microscopy data*, Nature Methods, 12 (11); 1065-1071 (2015).
[2] F. Levet et al. *A tessellation-based colocalization analysis approach for single-molecule localization microscopy.* Nature Communications (10), Article number: 2379 (2019).