Visual proteomics using whole-lamella 2D template matching

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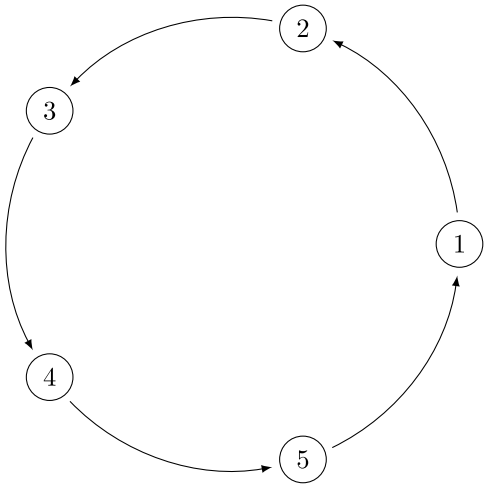
Abstract

Localization and characterization of biomolecules inside a cell is the fundamental quest of all biological imaging. Fluorescence microscopy can localize biomolecules inside whole cells and tissues, but its ability to count biomolecules and accuracy of the spatial coordinates is limited by the wavelength of visible light. Cryo-electron microscopy on the other hand provides highly accurate position and orientation information of biomolecules but is often limited to small fields of view inside a cell, providing only limited biological context. In this study we use a data-acquisition scheme called “Fast Observation of Whole Lamella” (FOWL) to collect cryo-electron microscopy data over thin central sections, representing roughly 1% of the total cellular volume, of neutrophil-like mouse cells. We use 2D-template matching to determine localization and orientation of the large subunit of the ribosome in these cross-sections. We furthermore use 2D-template matching to test for complex formation with the small ribosomal subunit and used relative orientations of ribosome to assign ribosomes to polysomes. Overall these results provide a “map” of translational activity in cross-sections of mammalian cells. We envision that using these high-throughput cryo-EM data collection and 2D template matching approaches will advance visual proteomics to complement other single-cell “omics” techniques, such as flow-cytometry and single-cell sequencing.

Introduction

Figures



References