Investigating Near Movement-Free Imaging Without GOLD

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Beam-induced movement (BIM) is long recognized as a significant problem in cryogenic electron microscopy (cryo-EM) and oftentimes limits achievable resolution. BIM is likely originated from pre-existing mechanical stress frozen into the sample that is released upon electron irradiation. Russo et al. proposed that BIM is caused by the buckling effect and subsequent deformation of the suspended ice layer during sample vitrification. The magnitude of BIM during the burst phase of sample movement depends on the shape and size of the ice layer set by the supporting foil of grids. HexAuFoil grids, a new recently developed sample support technology, seems promising to eliminate buckling of ice layer to reduce BIM to sub-angstrom. Conversely, this long-awaited gold grids might raise the following concerns to general users in the cryo-EM field, a) affordability, b) feasibility for vitrification with hole size as small as 200-nm, c) suitability for tilt image acquisition with 200-nm holes, and d) availability due to supply chain shortage. Here, we propose an alternative strategy to minimize BIM in cryo-EM using traditional Quantifoil grid through unique image acquisition scheme. We hypothesize that the global buckling effect occurred during vitrification could be partially released through pre-exposures at the site adjacent to the region of interest. Therefore, we quantify the BIM among the multi-shots within the same hole in a square. Quantifoil grids with various spacing were adopted to investigate the correlation between BIM pattern and the order of image acquisition within the same hole.
The finding of this study could enhance our understanding towards the underlying mechanism of BIM, thus providing alternative means to reach near movement-free imaging in cryo-EM.