IMPACT OF SPHERICAL ABERRATION ON STRUCTURED-ILLUMINATION MICROSCOPY

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The effect of depth-induced spherical aberration (SA) on structured illumination microscopy (SIM) [1] is investigated. A three-dimensional (3D) depth-variant (DV) imaging model was developed (by extending the model in [2]) to predict the grid images used in the SIM approach to estimate improved optical sections. The model incorporates SA due to imaging depth within a sample when there is a refractive index (RI) mismatch between the specimen and the immersion medium of the lens. It was implemented using a stratum-based model approximation and multiple depth-variant point-spread functions (PSFs) [3]. Simulations predicted by our model were computed and compared to images acquired from a 15 μm spherical shell.

Materials and Methods: The modulated images were acquired using the ApoTome attachment on a Zeiss Axio Imager microscope from a slide (Kit F-24634., Invitrogen Inc.) with 15-μm diameter polystyrene beads (RI = 1.59) embedded in optical cement (RI = 1.52 upon curing) using a 40x/1.3NA oil lens and a 20x/0.8NA lens using a wavelength of 519 nm. Simulations were obtained to match the imaging conditions using a Matlab implementation of the DV model.

Results: Simulated observations including SA predicted by our model capture the main features of the experimental data. Corresponding reconstructions calculated with traditional methods show undesirable artifacts (Fig. 1) and demonstrate the need for the development of a new computational approach for SIM reconstructions based on the DV imaging model presented here.

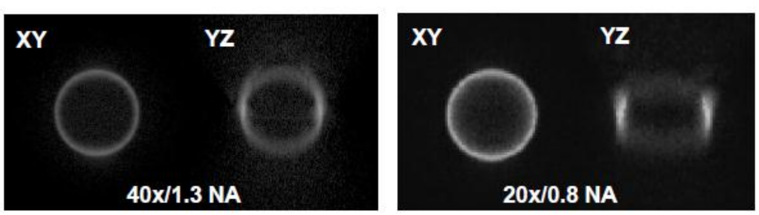

Figure 1: SIM reconstructions from measured images of a 15-μm diameter polystyrene bead.

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