Spherical Aberration Detection and Compensation in Widefield Fluorescence Microscope Images

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Practical flaws in microscope lens reduce the signal to noise ratio of the collected data and degrades the image quality. Spherical aberrations plays a significant role in this image degradation. We propose a new method for detecting spherical aberrations in an image, and to utilize it in deconvolution while taking advantage of both the parametric modeling of the PSF, polynomial fitting of the angle coefficient, and blind deconvolution.

Spherical aberration is caused by operating the microscope and lens outside the design conditions. Ideally, the lens immersion medium and sample embedding medium match, and the sample is on the surface of the cover-slip. Unfortunately, in reality, spherical aberration is the norm, rather than the exception. One of the primary causes of spherical aberration is a mismatch between the refractive index of the lens immersion medium and the embedding medium of the sample. Spherical aberration increases as you focus further into the sample, reduces the resolving power of the lens, and reduces the signal to noise ratio of the collected data. In this project we propose a new method for spherical aberration detection and compensation.

We show that a correlation between spherically aberrated PSFs and the XZ or YZ projection of the data can result in accurate estimation of the spherical aberrations coefficient. Then, we study the characteristic of spherically aberrated PSFs and show that spherical aberrations causes an increase in the image energy (which is not a function of the physical energy). We also show that completely compensating for spherical aberrations results in an image with minimum energy, given that everything else is fixed. Finally, we devise a scheme for detecting and correcting the spherical aberrations based on energy minimization.

We show that we can correctly detect the spherical aberrations coefficient in real biological images and in spherically aberrated beads and show the effect of this estimation process on the quality of results from the MLE-based blind deconvolution algorithm and on the Weiner filter with theoretically constructed PSF.

In order to correctly compensate for spherical aberrations, an accurate model of the PSF is needed. In the proposed PSF model we utilize a PSF equation that combines the theoretical modeling (Hopkins, 1955; Wilson, 1995) of the PSF and polynomial fitting of the angle factor from ground-truth values.