Blind Point Spread Function and Illumination in Image Scanning Microscopy

S. Labouesse[1], T. Mangeat[1], E. Premillieu[2], R. Piestun2[2]

[1] LITC Core Facility, Centre de Biologie Intégrative, Université de Toulouse, CNRS, UPS, 31062 Toulouse, France; [2] Department of Electrical, Computer and Energy Engineering, University of Colorado Boulder, Colorado, 80309, USA.

Abstract

Structured Illumination Microscopy (SIM) allows to double the resolution of a standard fluorescence microscope with low phototoxicity. SIM requires a precise knowledge of the illumination patterns, making it costly and difficult to calibrate. In many biological applications, the illuminations can be substantially modified once reaching the image plan due to the propagation in the sample itself. By using a blind approach, we greatly simplify the use of SIM and open the use of random illuminations (RIM), like speckle patterns [1,2,3]. This strategy allows super-resolution with low-cost instrumentation and is robust to optical aberrations in the excitation path. However, RIM remain sensible to aberrations in the collection path. Blind Image Scanning Microscopy (Blind-ISM) consider the collection point spread function (PSF) to be unknown as well as the illumination. Images are modeled as the convolution of a shift invariant PSF with the product of the sample with a single illumination shifted at multiple positions. Classical ISM [4], scan a focused excitation beam on the sample. In Blind-SIM, the unknown illumination can be any structured pattern, here we will consider a speckle pattern. A nonconvex optimization problem is solved with an iterative algorithm to jointly recover the PSF, the positions, the illumination and the sample. The scalar and shift ambiguity in the solution only allows relative positions and relative intensities estimations. We illustrate the performances of Blind-ISM in simulations in presence of strong aberrations and noise in terms of resolution and PSF estimation. Empirically we observe that the achieved resolution depends on the frequency content of the illumination and of the PSF. Super-resolution factor greater than 2 can be achieved with an evanescent speckle illumination.

References

1. Mudry, Emeric, et al. "Structured illumination microscopy using unknown speckle patterns." Nature Photonics 6.5 (2012): 312-315.

2. Idier, Jérôme, et al. "On the superresolution capacity of imagers using unknown speckle illuminations." IEEE Transactions on Computational Imaging 4.1 (2017): 87-98.

3. Mangeat, Thomas, et al. "Super-resolved live-cell imaging using random illumination microscopy." Cell reports methods 1.1 (2021): 100009.

4. Müller, Claus B., and Jörg Enderlein. "Image scanning microscopy." Physical review letters 104.19 (2010): 198101.