WAVELET BASED ADAPTIVE OPTICAL MICROSCOPY

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Aberrations severely limit the applicability of fluorescence microscopy when imaging deep inside tissue. Such disturbances are commonly caused by refractive index mismatches and heterogeneity in the distribution of the index of refraction within specimen. As a result, common microscopy techniques such as confocal microscopy can only operated effectively at limited focusing depths. Super-resolution methods, such as stimulated-emission-depletion (STED) microscopy, are even more sensitive to aberrations and so are limited to even shallower depths. To overcome these limitations one has to resort to adaptive optics [1].

A crucial issue that must be tackled in order to successfully deploy adaptive optics in these microscopes is the problem of measuring aberrations. Direct wavefront sensing techniques borrowed by the astronomy field have been considered, especially in the context of two-photon excitation microscopy [2]. Nevertheless such techniques have found limited adoption in some microscopy techniques, such as STED microscopy for example, since one must introduce additional equipment in the microscope and modify the sample preparation protocol.

A more versatile approach is to try to deduce the aberrations indirectly, by carefully analysing the images obtained with the microscope. When employing this approach, one commonly defines an image quality metric and suppresses the aberration by iteratively optimising the setting of a wavefront correction device, such as a deformable mirror or a spatial light modulator. Although this approach has been widely demonstrated in the past in a number of microscopy techniques, its accuracy is hampered by the presence of shot noise and by the unknown specimen structure present in the images. In this work we outline how one can use wavelets theory to successfully address these shortcomings.

We define novel wavelet-based metric optimization schemes for aberration correction and apply them to different high-resolution and super-resolution microscopes. This flexible approach is shown to be applicable across a range of specimens and imaging modalities.

[1] M. J. Booth, "Adaptive optical microscopy: the ongoing quest for the perfect image", Light Science Applications, **3**, e165 (2014).

[2] K. Wang, *et al.*, "Rapid adaptive optical recovery of optimal resolution over large volumes", Nature Methods **11**, 625–628 (2014).