

# NOISE PROPAGATION IN STRUCTURED ILLUMINATION MICROSCOPY

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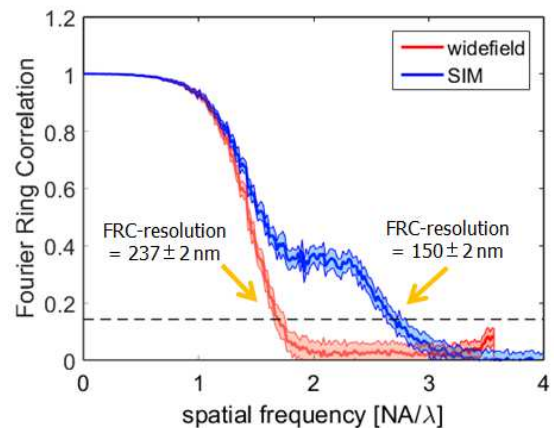
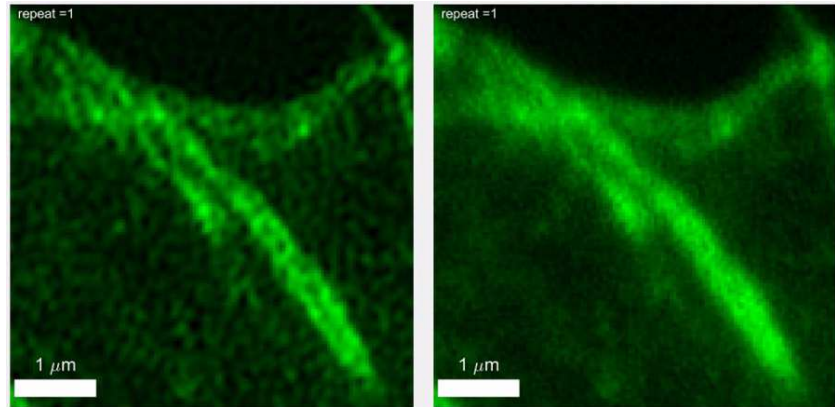
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Structured Illumination Microscopy (SIM) is an established super-resolution microscopy technique for providing an enhanced lateral resolution in combination with optical sectioning [1]. The default image reconstruction method in the field is so-called generalized Wiener filtering [2]. This SIM reconstruction technique suffers from a noise enhancement artefact, which manifests itself as a grainy haze that is added to the reconstructed image. We present a quantitative description of this artefact by tracking the effect of noise in the raw image acquisitions through all processing steps of the image reconstruction algorithm. It appears that the SIM noise artefact is due to selective amplification of noise in specific regions of spatial frequency space. We also propose a tailored version of the generalized Wiener filtering method that guarantees a flat noise spectrum. The analysis of noise propagation also entails a prediction for image quality and resolution measures such as the achievable Spectral Signal-to-Noise-Ratio (SSNR) [3] and the Fourier Ring Correlation (FRC) [4]. In the presentation we will describe simulations, theory and experimental results concerning our analysis of noise propagation, and we will provide an outlook to, in particular, a benchmark, based on the SSNR concept, of SIM with respect to other super-resolution techniques.

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*SIM reconstructions according to the state-of-the-art (top left) and according to the newly proposed reconstruction technique (top right) showing the removal of the typical SIM noise artefact. Measured FRC curves quantifying the resolution enhancement of SIM compared to widefield (bottom).*

[1] L. Schermelleh, R. Heintzmann, and H. Leonhardt, *J. Cell Biol.* Vol. 190, 165, 2012.

[2] M.G.L. Gustafsson et al., *Biophys. J.* Vol. 94, pp. 4957–4970, 2008.

[3] M. Unser et al., *Ultramicroscopy* Vol. 23, 39, 1987.

[4] R.P.J. Nieuwenhuizen et al., *Nature Methods.* Vol. 10, 557, 2013.