SUPER-RESOLUTION STOCHASTIC FLUCTUATION MICROSCOPY -
A COMPARISON BETWEEN SOFI AND STORM

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1. INTRODUCTION
Localization microscopy (e.g. PALM, STORM) achieves nanometer-scale resolutions by
sequentially imaging and localizing stochastically activated fluorophores. Super-resolution
optical fluctuation imaging (SOFI) likewise requires image sequences of independently
blinking emitters, but is based on the statistical analysis of fluorescence fluctuations and
enables super-resolution by computing higher-order temporal cumulants, or spatio-temporal
cross-cumulants [1].
In this work, we compare the two post-processing techniques used for STORM and SOFI and
identify their advantages and limitations [2].

2. SIMULATION & EXPERIMENT
In a Monte Carlo simulation, we generated image
sequences of randomly blinking fluorophores placed
arbitrarily on two parallel lines. We investigated the
effect of the rate ratio, the labeling density, the peak
signal-to-noise ratio (pSNR) and the line separation
distance on the relative visibility of the two lines.
For the experimental verification, we imaged BG-
Cy3-Cy5-labeled microtubule structures in human
osteosarcoma cells (U2OS) [3] and compared the
super-resolved images obtained by the SOFI and
STORM algorithms (Figure 1).

3. RESULTS & CONCLUSION
Although STORM achieves higher resolution enhancements, SOFI proves its potential as an
attractive, complementary alternative, because it works consistently over a wide range of
blinking statistics and tolerates much lower SNRs. Additionally, the different cumulant orders
can be used to determine the underlying molecular photo-switching statistics, which may be
useful in evaluating the applicability of STORM on a measured image sequence.

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Johnsson, K., and Lasser, T., “Targeted photoswitchable probe for nanoscopy of biological