

LIVE- AND FIXED-CELL SUPER-RESOLUTION MICROSCOPY ENABLED BY OPEN-SOURCE ANALYTICS IN IMAGEJ

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ImageJ is one of the main platforms for algorithms enabling super-resolution microscopy approaches that depend on an analytical step. In this tutorial, I will give an overview of how ImageJ-based image analysis is being employed to generate, qualify and quantify super-resolution microscopy data. I will then focus on super-resolution methods that are purely enabled by analysis of imaging data without the optical modification of microscopes, such as Single Molecule Microscopy methods (e.g.: PALM, STORM and DNA-PAINT) and the recently developed Super-Resolution Radial Fluctuations (SRRF) approach [1] (Fig. 1) developed by our laboratory. Throughout the tutorial, I will give walkthrough examples of how to acquire and analyse data with these methods, as well as discuss how to optimise image quality [2] and discuss pitfalls. The tutorial will be setup so that participants will be able to quickly translate the discussed approaches into their own research.

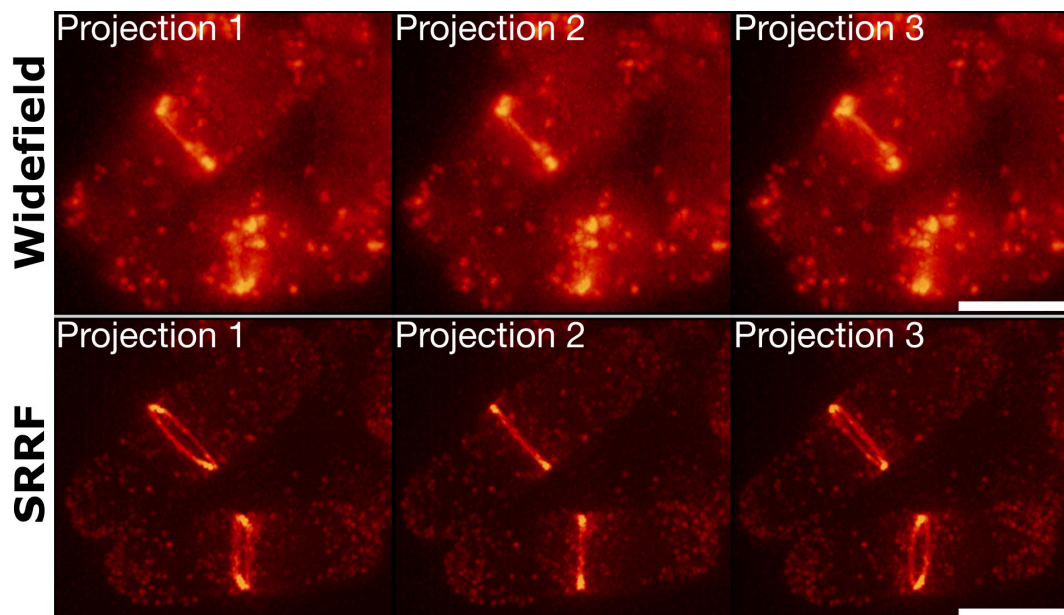


Fig 1. Three-dimensional structure of the stably-formed division ring in *S. pombe*. Widefield and SRRF imaging of LifeAct-GFP labelled actin division ring viewed from three different angles. Scale bars = 5 μ m.

1. Gustafsson, N., Culley, S., Ashdown, G., Owen, D. M., Pereira, P. M. & Henriques, R. Fast live-cell conventional fluorophore nanoscopy with ImageJ through super-resolution radial fluctuations. *Nat. Commun.* **7**, 12471 (2016).
2. Culley, S., Albrecht, D., Jacobs, C., Pereira, P. M., Leterrier, C., Mercer, J. & Henriques, R. NanoJ-SQUIRREL: quantitative mapping and minimisation of super-resolution optical imaging artefacts. *bioRxiv* (2017). doi:10.1101/158279