



## UKE Paper of the Month April 2023

### Expansion-enhanced super-resolution radial fluctuations enable nanoscale molecular profiling of pathology specimens

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[Nature Nanotechnology, 10. April 2023 Epub ahead of print](#)

#### ABSTRACT:

Expansion microscopy physically enlarges biological specimens to achieve nanoscale resolution using diffraction-limited microscopy systems. However, optimal performance is usually reached using laser-based systems (for example, confocal microscopy), restricting its broad applicability in clinical pathology, as most centres have access only to light-emitting diode (LED)-based widefield systems. As a possible alternative, a computational method for image resolution enhancement, namely, super-resolution radial fluctuations (SRRF), has recently been developed. However, this method has not been explored in pathology specimens to date, because on its own, it does not achieve sufficient resolution for routine clinical use. Here, we report expansion-enhanced super-resolution radial fluctuations (ExSRRF), a simple, robust, scalable and accessible workflow that provides a resolution of up to 25 nm using LED-based widefield microscopy. ExSRRF enables molecular profiling of subcellular structures from archival formalin-fixed paraffin-embedded tissues in complex clinical and experimental specimens, including ischaemic, degenerative, neoplastic, genetic and immune-mediated disorders. Furthermore, as examples of its potential application to experimental and clinical pathology, we show that ExSRRF can be used to identify and quantify classical features of endoplasmic reticulum stress in the murine ischaemic kidney and diagnostic ultrastructural features in human kidney biopsies.

#### STATEMENT:

*This interdisciplinary work expands the boundaries of experimental histopathology at the nanoscale across multiple clinical and scientific disciplines, helping to democratize and to integrate super-resolution microscopy, clinical and experimental pathology, thereby paving the way for the development of molecular nanopathology and standardized nanometrics to better diagnose, stratify and treat patients.*

#### BACKGROUND:

This work was lead and performed by the group of Prof. Puelles (III. Department of Medicine) in internal collaboration with the Hamburg Center for Kidney Health, Center for Molecular Neurobiology Hamburg, Departments of Biochemistry and Molecular Cell Biology, General, Visceral and Thoracic Surgery, Obstetrics and Fetal Medicine, Institutes of Pathology, Neuropathology, Mildred Scheel Cancer Career Center and the I. Department of Medicine. The first author, Dr. Kyliès is a clinician-scientist, focussed on kidney disease and the development of novel tissue analysis tools. Funding included the Collaborative Research Center 1192 "Immune-mediated kidney diseases" and the intramural clinician scientist program.