SPATIALLY-CONTROLLED ILLUMINATION COMBINED WITH RESCAN CONFOCAL MICROSCOPY TO REDUCE PHOTODAMAGE AND ENHANCE IMAGE QUALITY.

Venkataraman Krishnaswami\textsuperscript{a,b}, Giulia De Luca\textsuperscript{b}, Ronald Breedijk\textsuperscript{b}, Cornelis van Noorden\textsuperscript{a}, Erik Manders\textsuperscript{b}, Ron Hoebe\textsuperscript{a}

\textsuperscript{a}Cellular Imaging, Department of Medical Biology, Academic Medical Centre, University of Amsterdam, Amsterdam, The Netherlands; \textsuperscript{b}van Leeuwenhoek Centre for Advanced Microscopy, Swammerdam Institute for Life Sciences, University of Amsterdam, Amsterdam, The Netherlands.

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Fluorescence microscopy is an essential tool for live-cell imaging. However, excitation of fluorophores can induce photodamage. Hence, there is an inherent trade-off in fluorescence microscopy between image quality and photodamage. Recently, we introduced Re-scan Confocal Microscopy (RCM) as a new technology with improved signal-to-noise ratio compared to standard confocal microscopy, while reaching the highest possible confocal lateral resolution [1]. Earlier, we demonstrated that spatial control of illumination (SCIM) leads to reduced photodamage [2]. Here, we show that the combination of SCIM and RCM leads to reduced photodamage while preserving the and/or enhancing image quality. Implementation of spatially-controlled illumination in RCM uses a line scanning-based approach. Using information from previously acquired line images, the spatial illumination profiles for the upcoming lines is calculated by a prediction algorithm, during imaging. As a proof-of-principle, we show images comparing standard RCM to SCIM-RCM.
