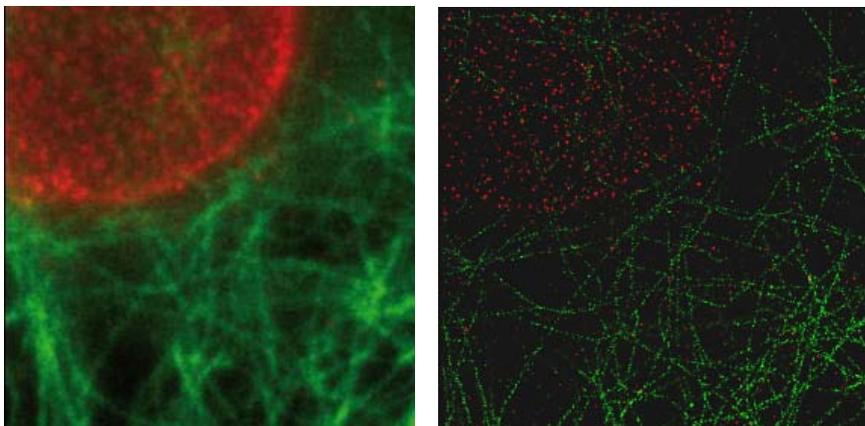


Leica Super-resolution Ground State Depletion (GSD): Breaking the barriers
Dr. Nathalie Garin
Leica Microsystems

The detailed knowledge of basic processes in life is a prerequisite to find the causes of previously incurable diseases and develop suitable therapies. GSDIM - **G**round **S**tate **D**epletion followed by **I**ndividual Single **M**olecule Return - enables the researcher to spatially localize proteins, other biomolecules or molecular processes in cells beyond the diffraction limit to gain insight into small functional structures.

The Ground State Depletion technology controls the emission of fluorochromes and localizes the position of single molecules down to a precision of 20nm. This allows reconstructing fluorescent images with a resolution far beyond the diffraction limit. With GSDIM, the fluorescent molecules in the specimen are almost completely switched off by driving them to the dark state using high power laser light. While most fluorescent molecules remain in the dark state, a small portion of molecules spontaneously return to the fluorescent state. This way, the signals of individual molecules can be acquired sequentially using a highly sensitive camera system and their spatial position in the specimen can be precisely measured and stored. Then, an extremely high-resolution image can be created from the position of many thousands of molecules. This enables to spatially separate intracellular structures that are situated very closely to one another and cannot be resolved using conventional fluorescence microscopy and sharply reproduced in an image.

One advantage of the Leica SR GSD system is that it uses conventional fluorescence markers such as fluorescent proteins, AlexaFluor® dyes or rhodamines to image within the cells with a resolution ten-times higher than diffraction limited microscopy.



Ptk2 cells, NPC staining: anti-NUPI53/Alexa532; Mt staining: anti- α tubulin/Alexa488. Courtesy: Wernher Fouquet, Leica Microsystems in collaboration with Anna Szymborsak and Jan Ellenberg, EMBL, Heidelberg, Germany