Discovery KitTM: dSTORM in cells

Prepare for super-resolution



Product Specifications

About

The ONI Discovery Kit™ for dSTORM imaging provides a modular workflow for immunofluorescent labeling of targets in cultured cells, which allows you to confidently detect extra and intracellular proteins in two channels with 20 nm resolution using dSTORM super-resolution microscopy. Two kit versions are available to provide the flexibility to optimize sample preparation steps, such as fixation, for you to label your targets of interest in your own samples.

Key Features & Benefits

50 samples with 1 kit

Volume provided is for 100 μL per sample. Protocol validated on uncoated and poly-L-lysine coated surfaces.

Optimize fixation & labeling for your targets

Use either the Standard Discovery Kit or the Discovery Kit with Strong Fixtive to fix and label your intra or extracellular targets of interest across a range of cell lines.

Flexibility to choose secondary probes for 2-color imaging

Highly specific anti-mouse, rabbit and rat secondary antibodies conjugated with best-in-class dSTORM fluorophores. Choose two secondary antibodies from a total of six to detect your own primary antibodies - three different species conjugated to either CF583R or AZ647. AZ647 is the same molecule as Alexa Fluor 647™.

dSTORM imaging using the Nanoimager

Stained cellular targets, including those expressed at low levels, can be detected using the Nanoimager, ONI's desktop super-resolution microscope, which allows simultaneous or sequential imaging of targets stained with CF583R and AZ647 secondary antibodies.

Analyze dSTORM with ONI's CODI platform

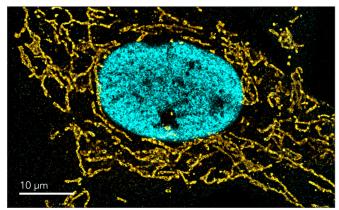
Extracting nanoscale morphological information from the stained structures is made possible by our cloud-based analysis platform, CODI, which includes clustering and counting tools to assess protein distribution in cells.

Validated across a range of proteins and cell types

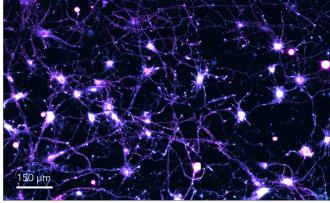
ONI's in-house team of experts has tested a range of protein targets with the dSTORM Discovery Kit, including:

- Intracellular organelles: mitochondria early and late endosomes, lysosomes
- Structural proteins: tubulin
- Nuclear proteins: histones, lamina
- Surface receptors including those at low expression: CD3, CD28, PD1

Tested in a range of cell types including human cancer cell lines, primary neurons, T cells, and mouse cell lines.



Histone H3K27ac (cyan) and mitochondria TOMM20 (gold) labeled with anti-rabbit CF583R and anti-mouse AF647 in U2OS cells.



Neuronal synaptic markers Bassoon (magenta) and Homer-1 (cyan) labeled with anti-mouse AZ647 and anti-rabbit CF583R (cyan) in primary rat cortical neurons.



learn more oni.bio/dstormdiscoverykit

Kit Versions



dSTORM Discovery Kit

The standard dSTORM Discovery Kit helps you prepare cells for dSTORM imaging with ONI's optimized reagents and protocols, allowing you to save time and obtain robust super-resolution data from your targets of interest in a fast and simplified manner. We recommend this kit for researchers studying intracellular non-membrane proteins. Exceptions are structural proteins, such as tubulin and actin, which are optimally fixed with the stronger fixative.

PFA-based fixative: Ready-to-use PFA is provided in a tear-off amber glass vial to avoid polymerization or deterioration. Extra PFA is provided for a post-fixation step so antibody labeled samples can be stably stored in the fridge.

Blocking buffers: Simultaneous blocking and permeabilization to save time and excellent permeabilization all the way to the nucleus.

Fluorescently-labeled F(ab')2 secondary antibodies:

Smaller than full length IgG to reduce linkage error and labeled with CF583R and AZ647, the best fluorophores for dSTORM for 2-channel imaging. Choose two antibodies from three different options (rat, mouse or rabbit) with either CF583R or AZ647.

BCubed imaging buffer: Our dSTORM imaging buffer provides excellent blinking and anti-bleaching activity.

Comprehensive protocol: Guides you through sample preparation, includes troubleshooting notes and advice to select and optimize antibodies against your protein of interest.



dSTORM Discovery Kit with Strong Fixative

PFA can be insufficient to cross-link many membrane proteins, which remain mobile¹. This can lead to antibody-induced protein clustering^{2,3}. The dSTORM Discovery Kit with strong fixative immobilizes membrane proteins using a combination of PFA and glutaraldehyde. This enables you to confidewntly measure membrane protein clustering. Because glutaraldehyde can disrupt some epitopes this kit provides both fixatives for you to compare and choose the best fixative for your protein of interest.

PFA-based fixative and Fixation Supplement for strong fixation: Ready-to-use PFA is provided in a tear-off amber glass vial to avoid polymerization or deterioration. Extra PFA is provided for a post-fixation step so antibody labeled samples can be stably stored in the fridge. The Fixation Supplement (glutaraldehyde-based) helps minimize residual mobility and avoid artifacts.

Quenching buffer: A quenching reagent is supplied to fully reduce the glutaraldehyde to minimize autofluorescence and non-specific antibody binding.

Included: Fluorescently-labeled F(ab')2 secondary antibodies, Optimized blocking and permeabilization buffers, BCubed imaging buffer, and Comprehensive protocol

- 1. Tanaka, K., Suzuki, K., Shirai, Y. et al. Membrane molecules mobile even after chemical fixation. Nat Methods. 2010; 7, 865-866. https://doi.org/10.1038/nmeth.f.314.
- Stanly TA, Fritzsche M, Banerji S, García E, Bernardino de la Serna J, Jackson DG, Eggeling C. Critical importance of appropriate fixation conditions for faithful imaging of receptor microclusters. Biol Open. 2016; 5(9): 1343-50. doi: 10.1242/bio.019943.
- 3. Werner C, Sauer M and Geis C. Super-resolving Microscopy in Neuroscience. Chem. Rev. 2021; 121 (19): 11971–12015.







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