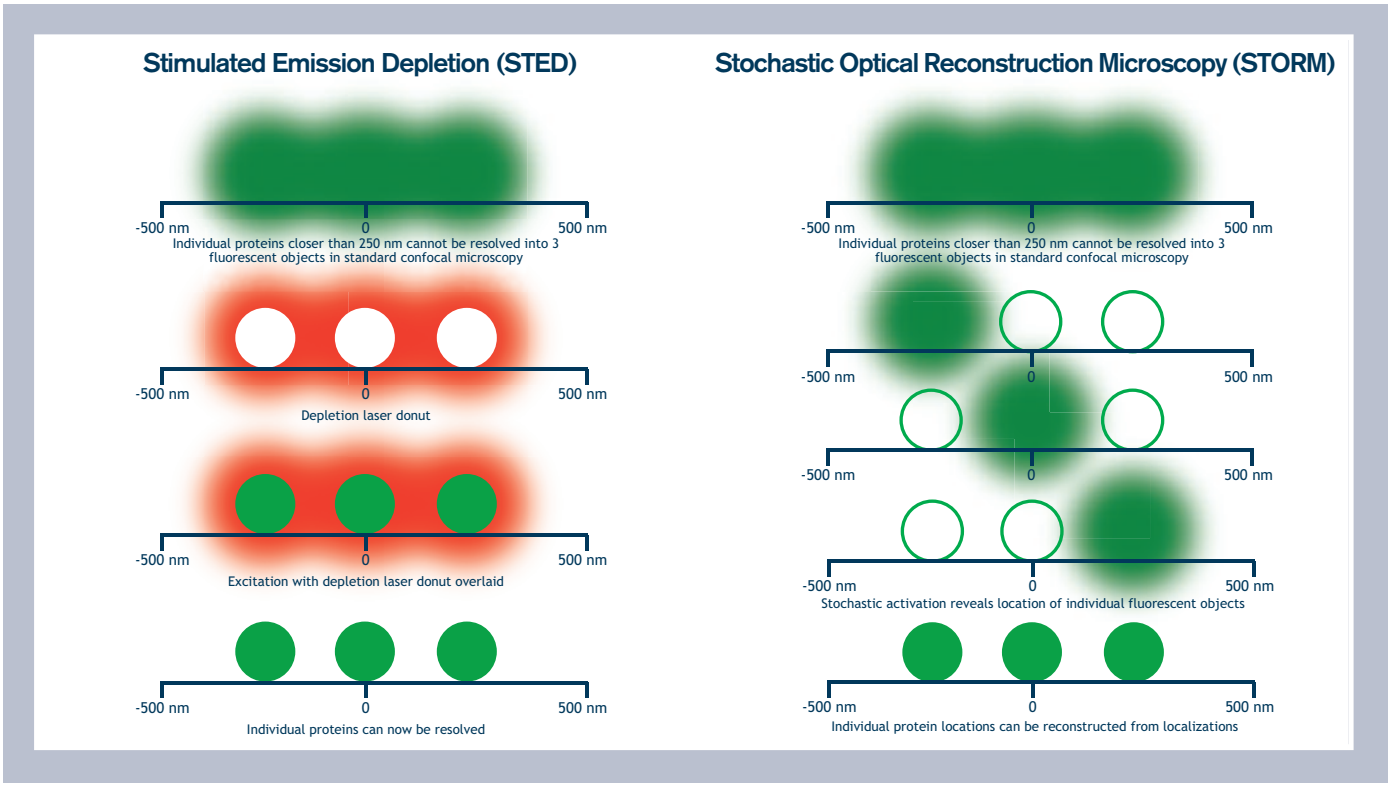
Jackson ImmunoResearch LABORATORIES, INC.

Specializing in **Secondary Antibodies** and **Conjugates**

Secondary Antibody Conjugates for **Superresolution Microscopy**

Fluorescence microscopy has become the workhorse imaging tool for biologists studying cellular structure and mechanism at the submicron scale. In practicality, for standard confocal microscopy experiments, objects closer than 250 nm in the lateral plane, and 500 nm in the axial direction cannot be discerned, and this limits the microscopist from a wealth of potential biological information.

Recently however, significant breakthroughs have been made in fluorescence microscopy. It is now possible to resolve cellular components in the range of 10 to 30 nanometers in the lateral, and 50-60 nm in the axial planes. Techniques such as Stimulated Emission Depletion (STED) and Stochastic Optical Reconstruction Microscopy (STORM) for example, have conquered the diffraction barrier and are now deepening what can be visualized at the microscopic level. **Researchers Eric Betzig, Stefan W. Hell, and William E. Moerner received the 2014 Nobel Prize in chemistry for these breakthroughs.**



Jackson ImmunoResearch offers a selection of labeled secondary antibodies with dyes known to be effective in STED, STORM, and related superresolution microscopy methods.

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Antibody Conjugate Fluorophores for STED

A stimulated emission depletion experiment produces superresolution images by narrowly confining the fluorescing region of a sample. To do so, it utilizes a depletion and an excitation laser to illuminate a very small region at the center of a "donut" formed by the two lasers. Dyes suited to STED experiments must have a high emission cross section with the STED laser wavelength and efficiently achieve a high saturation. They should have a low propensity for photo-bleaching, have high quantum yields and contrast, and contain sufficient density of labeling in close proximity to the target.

STED		Excitation (nm)	Emission (nm)
Alexa Fluor® 488		493	519
Fluorescein / FITC		492	520
Alexa Fluor® 594		591	614

Reference: Farahani, J. N. *et al*, Stimulated Emission Depletion (STED) Microscopy: from theory to practice. Microscopy: *Science Technology, Applications and Education.* (2010) 1539-1547.

Antibody Conjugate Fluorophores for Single Molecule Localization Experiments

For STORM applications both activation and emission dyes are on the same antibody. Jackson ImmunoResearch provides secondary antibodies conjugated to dyes that can "self-switch", resulting in high quality images without the need for dual-labeled antibodies. This was demonstrated in 2008 by Heilemann *et al.* and was referred to as direct STORM (dSTORM). The best dyes for single molecule localization are typically very bright and result in enough photons to reliably produce tight Gaussian distributions.

STORM		Excitation (nm)	Emission (nm)
Alexa Fluor® 488		493	519
Alexa Fluor® 647		651	667
Cy5		650	670

Reference: Dempsey *et al*, Evaluation of fluorophores for optimal performance in localization-based super-resolution imaging. *Nature Methods* **8** (2011) 1027-1036.

Certified by BSI to ISO 9001:2015 under certificate number FM 545248.

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