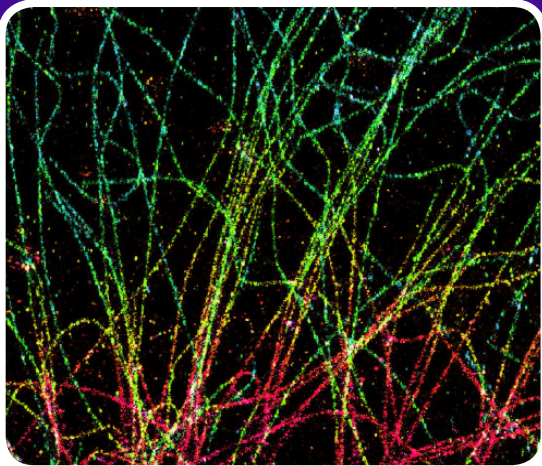


# Super-Resolution Microscopy Dyes



## CF® Dyes for multicolor super-resolution microscopy

Recent publications comparing synthetic dyes for super-resolution imaging have shown that CF® dyes give the best performance for multiple methods. The superior brightness, photostability, and photochemical switching properties of certain CF® dyes are ideal for 3-D SIM, 3-D STORM, and other super-resolution techniques. Biotium's CF®405M has been found to be the brightest and most photostable short wavelength fluorescent dye for SIM. Several CF® dyes spanning the visible red, far-red, and near-infrared spectra have been validated for STORM, including three color imaging with CF®568, CF®647, and CF®680. See Lehmann et al. 2015, and a full list of references on the next page. Biotium offers a wide selection of CF® dye labeled antibodies, including single-label secondaries with low degree of labeling, which is reported to be optimal for STORM<sup>1</sup>. We also offer other conjugates, reactive dyes, and labeling kits.

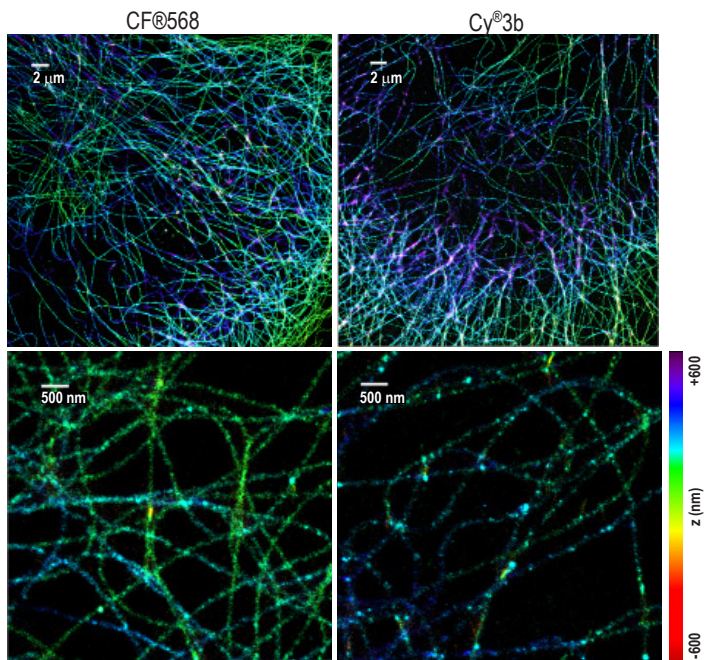


Figure 1. CF®568 (left) produces better images than Cy®3b (right) in 3-D STORM microscopy. Fixed cells were stained with mouse anti-tubulin antibody followed by dye-conjugated anti-mouse secondary antibodies. See Figure 2 legend for imaging conditions. Dye molecules were photoswitched and imaged using a 560 nm laser; a 405 nm laser was used to assist dye reactivation to the emitting state.

## CF® dyes and related products

- Primary antibody conjugates
- Labeled secondary antibodies
- Single-label secondaries for STORM
- Phalloidin and other bioconjugates
- Mix-n-Stain™ antibody labeling kits
- Protein labeling kits
- Full selection of reactive dyes
- Dyes for bioorthogonal labeling
- Small ligand (SNAP-tag®, CLIP-tag™, HALO-tag®) labeling kits

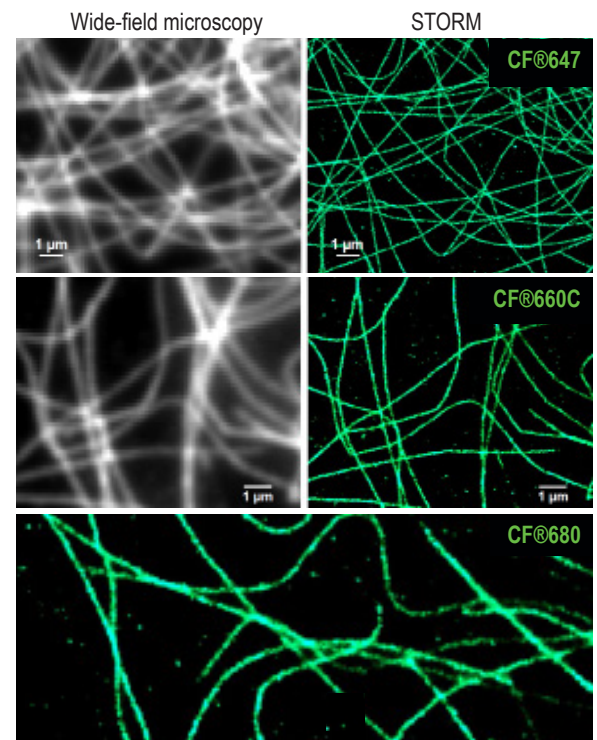


Figure 2. Comparison of conventional wide-field microscopy (left) with STORM (right) using CF® dye conjugates. Fixed cells were stained with mouse anti-tubulin antibody followed by CF® dye conjugated anti-mouse secondary antibody (top row: CF®647, middle row: CF®660C, bottom row: CF®680). For STORM, samples were sealed in buffer that contained 5% (w/v) glucose, 100 mM cysteamine, 0.8 mg/mL glucose oxidase, and 40 µg/mL catalase, in Tris-HCl (pH 7.5). Samples were imaged on a Nikon Ti-Eclipse w/ PFS microscope with a CFI Plan Apo Lambda 100x oil objective. Dye molecules were photoswitched and imaged using a 647 nm laser; a 405 nm laser was used to assist dye reactivation to the emitting state. Emission was collected with an Andor iXon Ultra 897 EMCCD camera for a total of 100,000 frames per image at a frame rate of 110 Hz.

<sup>1</sup> Bittel, A.M. et al. (2015). Proc. SPIE 9331, [Single Molecule Spectroscopy and Superresolution Imaging VIII, 93310M](https://doi.org/10.1117/12.2083209). doi: 10.1117/12.2083209  
Microscopy images courtesy of Sam Kenny and Professor Ke Xu, College of Chemistry, University of California, Berkeley.

## Super-resolution imaging techniques validated for CF® dyes

CF® Dye	Abs/Em maxima	Extinction coefficient	Super resolution application	References
CF@405S	404/431 nm	33,000	SIM	Demmerle et al. (2017). <a href="#">Nature Protocols 12, 988–1010</a> Essig et al. (2017). <a href="#">Immunity https://doi.org/10.1016/j.immuni.2017.11.008</a>
CF@405M	408/452 nm	41,000	SIM, STED	Kraus, F. et al. (2017). <a href="#">Nat Protoc 12, 1011-1028, doi:nprot.2017.020</a> (SIM) Markaki, Y. et al. (2013). <a href="#">Methods Mol Biol 950, 43-64</a> (SIM) Miron, E. et al. (2016). In: Mark C. Leake (ed.), <a href="#">Methods in Molecular Biology, vol. 1431, 127-140</a> (SIM) Ohgomi, T. et al. (2017). <a href="#">Eur J Neurosci 46, 2001-2014, doi:10.1111/ejn.13650</a> (SIM) Zhang, R. et al. (2019). <a href="#">bioRxiv 586461, doi: https://doi.org/10.1101/586461</a> (STED)
CF@488A	490/515 nm	70,000	STED, TIRF	Angelov, B. & Angelova, A. (2017). <a href="#">Nanoscale 9, 9797-9804, doi:10.1039/c7nr03454g</a> (STED) Zanetti-Domingues, L.C. et al. (2013). <a href="#">PLoS ONE 8(9): e74200</a> (TIRF)
CF@535ST	535/568 nm	95,000	STORM	Collaborator communication; contact techsupport@biotium.com for more information.
CF@555	555/565 nm	150,000	Multicolor STORM	Lehmann, M. et al. (2015). <a href="#">J Biophotonics DOI 10.1002/jbio.201500119</a>
CF@568	562/583 nm	100,000	Multicolor STORM, SIM, TIRF	Gong, Y.-N. et al. (2017). <a href="#">Cell Cycle, 1-13, doi:10.1080/15384101.2017.1371889</a> (STORM) Gorur, A. et al. (2017). <a href="#">J Cell Biol 216, 1745-1759, doi:10.1083/jcb.201702135</a> (STORM) Heller, J. (2017). <a href="#">OM&amp;P 3, 48-58, doi:doi:10.20388/omp2017.002.0045</a> (STORM) Jorgans, D.M. et al. (2017). <a href="#">J Cell Sci 2017 130: 177-189, doi: 10.1242/jcs.190967</a> (STORM) Karanasios, E. et al. (2016). <a href="#">Nat Commun 7: 12420, DOI: 10.1038/ncomms12420</a> (STORM) Kraus, F. et al. (2017). <a href="#">Nat Protoc 12, 1011-1028, doi:nprot.2017.020</a> (SIM) Lehmann, M. et al. (2015). <a href="#">J Biophotonics DOI 10.1002/jbio.201500119</a> (STORM) Lim, A. et al. (2017). <a href="#">Mol Biol Cell doi:mbc.E16-12-0820</a> (SIM) Turkowsky, B. et al. (2016). <a href="#">Anal Bioanal Chem DOI 10.1007/s00216-016-9781-8</a> (STORM) Zanetti-Domingues, L.C. et al. (2013). <a href="#">PLoS ONE 8(9): e74200</a> (TIRF) Zhang, M. et al. (2015). <a href="#">eLife 2015:10.7554/eLife.11205</a> (STORM)
CF@594ST	593/614 nm	115,000	STORM	Collaborator communication; contact techsupport@biotium.com for more information. <b>Note:</b> CF@594ST is a unique dye designed for STORM. Our original CF@594 is not suitable for STORM.
CF@633	630/650 nm	100,000	FIONA, gSHRImP, Single molecule tracking, TIRF	Bosch, P. J. et al. (2014). <a href="#">Biophys J 107, 803-814</a> (TIRF) Huang, T. et al. (2018). <a href="#">Biophysical Journal 114, 301–310</a> . (Single Molecule Tracking) Kim, H. J., and Selvin, P. R. (2013). <a href="#">SpringerReference Encyclopedia of Biophysics</a> (FIONA) Simonson, P. D. et al. (2011). <a href="#">Nano Lett 11, 5090-5096, DOI:10.1021/nl203560r</a> (gSHRImP) Zanetti-Domingues, L.C. et al. (2013). <a href="#">PLoS ONE 8(9): e74200</a> (TIRF) Zhang, R. et al. (2017). <a href="#">eLife 2017:6:e30959</a> (TIRF)
CF@640R	642/662 nm	105,000	FLImP, SIM, TIRF	Bosch, P. J. et al. (2014). <a href="#">Biophys J 107, 803-814</a> (TIRF) Loh, L. N. (2017). <a href="#">MBio 8, doi:mBio.02030-16</a> (SIM) Martin-Fernandez, M. L. et al. (2013). <a href="#">J Microsc 252, 16-22</a> (TIRF) Needham, S.R. et al. (2015). <a href="#">Biochem Soc Trans 43, 309–314</a> (FLImP) Needham, S.R. et al. (2016). <a href="#">Nat Commun 7, 13307, doi:ncomms13307</a> (FLImP) Zanetti-Domingues, L.C. et al. (2013). <a href="#">PLoS ONE 8(9): e74200</a> (TIRF) Zanetti-Domingues, L.C. et al. (2015). <a href="#">Prog Biophys Mol Biol, doi:S0079-6107(15)00047-4</a> (FLImP)
CF@647	650/665 nm	240,000	Multicolor STORM	Gong, Y.-N. et al. (2017). <a href="#">Cell Cycle, 1-13, doi:10.1080/15384101.2017.1371889</a> Lehmann, M. et al. (2015). <a href="#">J Biophotonics DOI 10.1002/jbio.201500119</a> Olivier, N. et al. (2013). <a href="#">Biomed Opt Express 4, 885-899</a> Turkowsky, B. et al. (2016). <a href="#">Anal Bioanal Chem DOI 10.1007/s00216-016-9781-8</a>
CF@660C	667/685 nm	200,000	Multicolor STORM	Turkowsky, B. et al. (2016). <a href="#">Anal Bioanal Chem DOI 10.1007/s00216-016-9781-8</a> Zhang, Z., et al. (2015). <a href="#">Nature Methods doi:10.1038/nmeth.3528</a>
CF@680	681/698 nm	210,000	Dual-Color 3D SMLM, Multicolor STORM	Früh, S.M. et al. (2015). <a href="#">Nature Communications 6, 7275</a> (STORM) Glebov, O.O. et al. (2017). <a href="#">Cell Rep 18, 2715-2728, doi:S2211-1247(17)30279-6</a> (STORM) Gorur, A. et al. (2017). <a href="#">J Cell Biol 216, 1745-1759, doi:10.1083/jcb.201702135</a> (STORM) Lehmann, M. et al. (2015). <a href="#">J Biophotonics DOI 10.1002/jbio.201500119</a> (STORM) Platonova, E. et al. (2015). <a href="#">ACS Chem Biol 10(6), 1411–1416</a> (STORM) Platonova, E. et al. (2015). <a href="#">Methods doi: http://dx.doi.org/10.1016/j.ymeth.2015.06.018</a> (STORM) Salvador-Gallego, R. et al. (2016). <a href="#">EMBO J. DOI 10.15252/embj.201593384</a> (STORM) Shrestha, R. L. et al. (2017). <a href="#">Nat Commun 8, 150, doi:10.1038/s41467-017-00209-z</a> (STORM) Turkowsky, B. et al. (2016). <a href="#">Anal Bioanal Chem DOI 10.1007/s00216-016-9781-8</a> (SMLM) Winterflood, C.M. et al. (2015). <a href="#">Biophys J, 109, 3–6</a> (SMLM) Zhang, Z., et al. (2015). <a href="#">Nature Methods doi:10.1038/nmeth.3528</a> (STORM)
CF@680R	680/701 nm	140,000	Multicolor STORM, Single-molecule spectroscopy, Single molecule tracking, STED	Conley, G. et al. (2017). <a href="#">Science Advances 3(10), e1700969 DOI:10.1126/sciadv.1700969</a> (STORM) Görlitz, F. et al. (2014). <a href="#">Progress Electromagnetics Res 147, 57-68</a> (STED) Huang, T. et al. (2018). <a href="#">Biophysical Journal 114, 301–310</a> (Single Molecule Tracking) König, I. et al. (2015). <a href="#">Nature Methods doi:10.1038/nmeth.3475</a> (Single molecule spectroscopy) Winter, F.R. et al. (2017). <a href="#">Scientific Reports 7, 46492, DOI: 10.1038/srep46492</a> (STED)
CF@750	755/777 nm	250,000	STORM	Collaborator communication; contact techsupport@biotium.com for more information.

FIONA: Fluorescence Imaging with One Nanometer Accuracy; FLImP: Fluorophore localization imaging with photobleaching; SHRImP: Single-molecule high-resolution imaging with photobleaching; SIM: Structured illumination microscopy; SMLM: Single molecule localization microscopy; STED: Stimulated emission depletion; STORM: Stochastic optical reconstruction microscopy; TIRF: Total internal reflection fluorescence

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