

CF™ Dyes

Next Generation Fluorescent Dyes

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CF™ Dyes Quick Reference Table

Visible spectrum

Far-red

Near-infrared

CF™ dye	Page	λ_{Ex} (nm)	λ_{Em} (nm)	Excitation *	Replacement for	Features and applications
CF™350	5	347	448	UV	Alexa Fluor® 350, AMCA, DyLight® 350	<ul style="list-style-type: none"> Brightest blue fluorescent conjugates for 350 nm excitation Highly water-soluble and pH insensitive
CF™405S	5	404	431	405 nm	Alexa Fluor® 405, Cascade Blue®, DyLight® 405	<ul style="list-style-type: none"> Better compatibility with common instruments
CF™405M	5	408	452	405 nm	BD Horizon™ V450, eFluor® 450, Pacific Blue®	<ul style="list-style-type: none"> More photostable than Pacific Blue® dye with less green spill-over Excellent choice for super-resolution imaging by SIM**
CF™405L	6	395	545	405 nm	Pacific Orange®	<ul style="list-style-type: none"> 405 nm excitable orange fluorescent dye for multicolor detection
CF™430	6	426	498	405 nm	Pacific Green®, BD Horizon™ V500, Krome Orange™	<ul style="list-style-type: none"> Photostable 405 nm excitable green dye Perfect match for the CFP filter set
CF™440	6	440	515	405 nm	Alexa Fluor® 430	<ul style="list-style-type: none"> Photostable 405 nm excitable green dye
CF™450	7	450	538	405 nm	Unique dye	<ul style="list-style-type: none"> Green dye with unique spectral properties
CF™488A	7	490	515	488 nm	ATTO 488, Alexa Fluor® 488, Cy®2, DyLight® 488, FAM, FITC, Fluorescein	<ul style="list-style-type: none"> Less non-specific binding and less red spill-over than Alexa Fluor® 488 Very photostable Compatible with super-resolution imaging by TIRF**
CF™514	8	516	548	488 nm	Alexa Fluor® 514	<ul style="list-style-type: none"> Green dye that can be separated from CF™488A by spectral unmixing
CF™532	9	527	558	532 nm	Alexa Fluor® 532, ATTO 532	<ul style="list-style-type: none"> Significantly brighter than Alexa Fluor® 532
CF™535ST	9	535	568	532 nm	Unique dye for STORM**	<ul style="list-style-type: none"> Orange dye designed for STORM super-resolution microscopy**
CF™543	10	541	560	532, 543, or 546 nm	Alexa Fluor® 546, Tetramethylrhodamine (TAMRA)	<ul style="list-style-type: none"> Significantly brighter than Alexa Fluor® 546
CF™555	10	555	565	532, 543, 546, 555, or 568 nm	Alexa Fluor® 555, ATTO 550, Cy®3, DyLight® 549, TRITC	<ul style="list-style-type: none"> Brighter than Cy®3 Validated in multicolor super-resolution imaging by STORM**
CF™568	11	562	583	532, 543, 546, 555, or 568 nm	Alexa Fluor® 568, ATTO 565, Rhodamine Red	<ul style="list-style-type: none"> Optimized for the 568 nm line of the Ar-Kr mixed-gas Brighter and more photostable than Alexa Fluor 568 Compatible with TIRF and multicolor STORM**
CF™594	12	593	614	532, 543, 546, 555, or 568 nm	Alexa Fluor® 594, ATTO 594, DyLight® 594, Texas Red®	<ul style="list-style-type: none"> Yields the brightest conjugates among spectrally similar dyes Extremely photostable
CF™594ST	12	593	614	532, 543, 546, 555, or 568 nm	Unique dye for STORM**	<ul style="list-style-type: none"> Specifically designed for super-resolution imaging by STORM**
CF™620R	13	617	639	633 or 635 nm	LightCycler® Red 640	<ul style="list-style-type: none"> Highly fluorescent dye with unique spectral properties
CF™633	14	630	650	633 or 635 nm	Alexa Fluor® 633, Alexa Fluor® 647, Cy®5, DyLight® 633	<ul style="list-style-type: none"> Yields the brightest antibody conjugates among spectrally similar dyes Far more photostable than Alexa Fluor® 647 Compatible with super-resolution TIRF, FIONA, and gSHRIMP**
CF™640R	15	642	662	633, 635, or 640 nm	Alexa Fluor® 647, ATTO 647N, Cy®5, DyLight® 649	<ul style="list-style-type: none"> Has the best photostability among dyes with Cy®5-like spectra Yields highly fluorescent protein conjugates Compatible with TIRF and FLIMP super-resolution techniques**
CF™647	16	650	665	633, 635, or 640 nm	Alexa Fluor® 647, ATTO 647N, Cy®5, DyLight® 649	<ul style="list-style-type: none"> Brighter than Cy®5 Compatible with multicolor super-resolution imaging by STORM**
CF™660C	17	667	685	633, 635, or 640 nm	Alexa Fluor® 660	<ul style="list-style-type: none"> Much brighter and more photostable than Alexa Fluor® 660 Compatible with multicolor super-resolution imaging by STORM**
CF™660R	17	663	682	633, 635, or 640 nm	Alexa Fluor® 660	<ul style="list-style-type: none"> Brighter than Alexa Fluor® 660 The most photostable 660 nm dye
CF™680	18	681	698	680 or 685 nm	Alexa Fluor® 680, Cy®5.5, DyLight® 680, IRDye® 680LT	<ul style="list-style-type: none"> The brightest among spectrally similar 680 nm dyes Validated in multicolor STORM and dual-color 3D super-resolution imaging** Compatible with LI-COR® Odyssey® System
CF™680R	18	680	701	680 or 685 nm	Alexa Fluor® 680, Cy®5.5, DyLight® 680, IRDye® 680LT	<ul style="list-style-type: none"> The most photostable 680 nm dye Suitable for labeling nucleic acids and small biomolecules Compatible with LI-COR® Odyssey® System Compatible with STED and single molecule spectroscopy**
CF™750	19	755	777	680 or 685 nm	Alexa Fluor® 750, Cy®7, DyLight® 750, IRDye® 750	<ul style="list-style-type: none"> Exceptionally bright and stable Highly water soluble without bearing excessive charge Validated in super-resolution imaging by STORM**
CF™770	19	770	797	785 nm	DyLight® 800, IRDye® 800CW, ZW800-1	<ul style="list-style-type: none"> Exceptionally bright and stable Compatible with LI-COR® Odyssey® System
CF™790	19	784	806	785 nm	Alexa Fluor® 790	<ul style="list-style-type: none"> Exceptionally bright and stable Highly water soluble without bearing excessive charge
CF™800	19	797	816	785 nm	Spectrally similar to Indocyanine green	<ul style="list-style-type: none"> Unique long wavelength near-infrared dye

*Visible and far-red dyes can be excited by a UV light source for epifluorescence microscopy.

**See pp. 20-21 for more information about CF™ dyes for super-resolution microscopy

CF™ Dyes Overview

Next-generation fluorescent dyes

CF™ dyes are a series of highly water-soluble fluorescent dyes spanning the visible and near-infrared (IR) spectrum for labeling biomolecules, especially proteins and nucleic acids. Developed by scientists at Biotium using new breakthrough chemistries, CF™ dyes rival or exceed the quality of other commercial dyes, such as Alexa Fluor® dyes, due to the following novel features.

Novel rhodamine chemistry

Rhodamine dyes are known for their excellent photostability and good fluorescence quantum yield; consequently several of the Alexa Fluor® dyes bear the rhodamine core structure. Unfortunately, traditional rhodamine chemistry makes it difficult to extend the fluorescence wavelength to the far-red region and even more challenging in the near-IR region, especially for water-soluble dyes for bioconjugation. Recently, Biotium scientists discovered a new way to prepare novel rhodamine dyes of any fluorescence color from green to near-IR. The new chemistry is a key element in the development of many of our CF™ dyes, particularly our far-red CF™ dyes, which are not only bright and water-soluble but also extremely photostable.

Excellent labeling efficiency

Reactive dyes for bioconjugation are generally susceptible to hydrolysis, which can cause problems for shipping, handling and storage and result in lower labeling efficiency. Heavily sulfonated dyes, such as the Alexa Fluor® dyes, DyLight® dyes and IRDyes® are particularly hygroscopic, worsening the hydrolysis problem. For example, the percent of active Alexa Fluor® 488 succinimidyl ester (SE) could be well below 50% by the time of application (according to Life Technologies' Alexa Fluor® 488 microscale labeling kit product information sheet, Invitrogen). In contrast, all of Biotium's amine-reactive CF™ dyes have a relatively stable form of SE, which is more resistant to hydrolysis than the SE in many of the Alexa Fluor® dyes. Accordingly, CF™ dye SE products generally give consistently higher labeling efficiency, thus providing users a better value.

Mix-n-Stain™ antibody labeling technology

Biotium has developed a breakthrough antibody labeling technology with CF™ dyes — Mix-n-Stain™ antibody labeling kits. With this technology, you merely need to mix your antibody with the reaction buffer and CF™ dye provided in the kit, and in 30 minutes you will have an optimally labeled CF™ dye-antibody conjugate ready for immunostaining. The labeling technology provides unprecedented convenience for antibody labeling. Mix-n-Stain™ labeled antibodies can be used for multicolor immunostaining, allowing staining with multiple primary antibodies from the same host species when pre-labeled primary antibodies are not available.

CF™ Dyes for Super-Resolution Microscopy

Recent publications comparing synthetic dyes for super-resolution imaging have shown 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 and single-molecule imaging techniques. See pp. 20-21 for more information.

Unrivaled near-infrared dyes

Near-IR dyes are typically much larger in size than dyes in the visible range. The large size often results in serious problems of low dye solubility, dye aggregation and poor fluorescence quantum yield. To overcome the problems, many commercial near-IR dyes, such as the near-IR Alexa Fluor® dyes, DyLight® dyes and IRDyes®, are prepared by placing a number of negatively charged sulfonate group on the dyes. While sulfonation improves dye solubility and fluorescence quantum yield to some degree, it creates another even more serious problem: non-specific binding of the bioconjugates prepared from the dyes. For example, conjugation to a highly negatively charged dye can dramatically alter an antibody's isoelectric point (iP), which is essential for maintaining specific antibody-antigen interaction. With this insight, Biotium scientists devised a revolutionary new approach to near-IR dye design that results in superior physical properties of the dyes without introducing an excessive amount of negative charge.

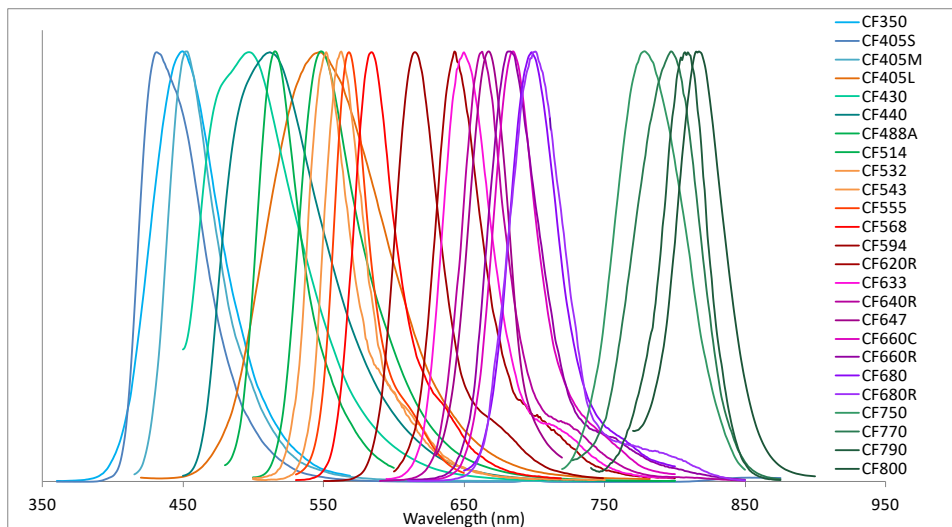
Biotium's near-IR CF™ dyes are based on the core structure of either cyanine dyes or rhodamine dyes. Those core structures are modified such that the intramolecular mobility of the dyes is restricted, which leads to higher quantum yield and better water solubility without adding excessive charge. As a result, near-IR CF™ dyes are much brighter and more photostable than any other near-IR dyes. Most importantly, antibodies labeled with near-IR CF dyes™ give far better signal-to-noise ratio in immunostaining compared with antibody conjugates prepared with other commercial near-IR dyes.

Multi-color flexibility

Biotium currently offers 26 CF™ dyes with additional colors in development. The CF™ dye product line includes reactive CF™ dyes, labeling kits, CF™-labeled secondary antibodies and streptavidin, and many other CF™-labeled biomolecules.

CF™ dyes and Mix-n-Stain antibody labeling technology are covered by pending U.S. and international patents. We welcome inquiries about licensing the use of our dyes, trademarks or technologies; email us at btinfo@biotium.com.

Emission spectra of CF™ dyes



CF™ Dyes

Frequently Asked Questions

Question	Answer
What does the CF in CF dyes stand for?	CF initially was an abbreviation for Cyanine-based Fluorescent dyes. These were the first patented CF dyes based on cyanine dye structures. Since then, our CF dye patent portfolio has expanded to include four different fluorescent dye core structures that cover the fluorescence spectrum from UV to NIR.
What are the chemical structures of CF dyes?	The exact chemical structures of CF dyes are currently confidential but will be fully disclosed at a later stage when pending patents become granted. In general terms, the structure of a CF dye may be divided into two parts: a) dye core structure (i.e. the aromatic ring skeleton that defines the dye's color or absorption/emission wavelengths), and b) core structure-modifying elements. At present, CF dyes bear the core structures of coumarin, pyrene, rhodamine or cyanine dyes. Blue fluorescent CF dyes are based on coumarin or pyrene dye core structure, while green to near-IR CF dyes are based on either cyanine or rhodamine dye core structures. Core structure-modifying elements refer to various chemical attachments to the core structure and are a key aspect of the CF dye invention that makes CF dyes superior to other commercial dyes.
What are the quantum yields of CF dyes?	The quantum yield of a fluorescent dye can vary widely depending on the dye's micro-environment if the dye is attached to a protein or other molecule. A good way to compare the relative quantum yields of different dyes is to plot the total fluorescence of the labeled proteins as a function of degree of labeling by the dyes, as we have done with CF dyes and other commercial dyes in the dye description pages in this guide.
How stable are CF dyes?	There are three aspects to dye stability: 1) chemical stability of the dye core structure; 2) stability of the reactive group; and 3) photostability of the dye. CF dyes bear the core structures of coumarin, pyrene, rhodamine or cyanine dyes, all of which are known to have excellent chemical stability. In general, the dyes are far more stable than the antibodies or other biomolecules they label. CF dyes are also stable enough for labeled nucleic acids to be used in PCR or nucleic acid hybridization, where high temperature is involved. Reactive CF dyes comprise a reactive group used in bioconjugation. Among the various reactive groups, only amine-reactive succinimidyl ester (SE) and thiol-reactive maleimide groups are susceptible to hydrolysis and therefore are moisture-sensitive. CF dye SE products are relatively more stable than other commercial SE dyes. This is because CF SE dyes are derived from aliphatic carboxylic groups, which results in a more stable SE form, while other commercial SE dyes usually are derived from aromatic carboxylic acid groups that yield a less stable SE form. Photostability refers to the dye's ability to withstand photobleaching. Photobleaching is mainly a concern when dyes are subjected to intense illumination for an extended period of time, such as during confocal microscopy. Among the four types of core structures, rhodamine is the most photostable, followed by cyanine, pyrene and coumarin cores. The structure-modifying groups and the way they are attached to the dye cores are a key innovative aspect of CF dye technologies that contributes to the superior photostability of CF dyes over that of other commercial dyes. In general, rhodamine-based CF dyes, whose wavelengths range from green to the near-IR region, offer the best photostability, making these dyes ideal for microscopy applications.
Are CF dyes sensitive to pH?	CF dyes are chemically stable within the pH range of at least 2–11. The fluorescence of most CF dyes is relatively insensitive to pH, except for that of CF405M, CF568, CF620R, and CF633. The fluorescence of these four CF dyes becomes weaker when pH drops below 4.5.
Are CF dyes fixable?	CF™ dyes can tolerate formaldehyde fixation. However, whether a CF™ dye-labeled probe is fixable will depend on the fixability of the probe itself. Proteins with free amine groups that bind other proteins generally are formaldehyde-fixable.
What is the difference between CF405s, CF405M, and CF405L?	All three of these dyes can be excited by the 405 nm laser (or UV mercury lamp). They differ in their emission wavelengths. CF™405S has the shortest blue fluorescence emission at 431 nm, while CF™405M has longer wavelength blue fluorescence emission at 452 nm. CF™405L has orange fluorescence emission at 545 nm. We recommend choosing the dye that best fits your instrument's detection settings (see pp. 5-6 for more information).
For several CF dye colors, there is an R form and a C form, both having similar absorption and emission spectra. In such a case, which of the two CF dyes should I choose?	Rhodamine-based CF dyes (designated R) generally have better photostability but weaker fluorescence than their cyanine-based equivalents (designated C). Therefore, rhodamine-based near-IR CF dyes are a better choice for microscopy, while cyanine-based CF dyes are more ideal for flow cytometry, Western blotting, and other applications where photobleaching is less of a concern. Another factor to consider is the size of the dyes. Some of the cyanine-based near-IR CF dyes are much larger than the rhodamine-based equivalents. For antibody labeling, either version of the CF dyes is suitable. However, for applications where the dye size may cause a steric problem, the smaller dye may be a better choice.
How soluble are CF dyes?	CF dyes are highly water soluble (>100 mg/mL). They are also very soluble in other polar solvents, such as DMSO, DMF, methanol and ethanol. However, CF dyes are poorly soluble or insoluble in non-polar solvents.
What are the charges on CF dyes?	Most CF dyes carry 1-2 negative charges while a few cyanine-based near-IR CF dyes carry 3-4 negative charges. However, the more negatively charged CF dyes comprise unique structural features that shield the negative charges such that the biomolecules (such as antibodies) the dyes label do not lose specificity due to the excessive negative charges.
Can CF dyes be used for STORM?	Several CF dyes have been validated in super-resolution imaging by STORM, as well as other super-resolution techniques. Biotium also offers dyes specifically designed for STORM imaging. See pp. 20-21 for more information.
What are the major applications of CF dyes?	CF dyes are ideal for protein labeling because of their high water solubility, which reduces fluorescence quenching. They are also useful for labeling oligonucleotides that require multiple copies of a dye for maximal fluorescence, such as the preparation of FISH probes, where water soluble dyes can minimize fluorescence quenching. Finally, CF dyes make excellent polar tracers that can be used for visualizing the morphology or long-term tracing of neurons.

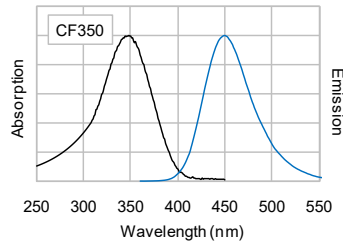
CF™ 350

A bright UV-excitable blue fluorescent dye

Technical Summary

Abs/Em Maxima: 347/448 nm
Extinction coefficient: 18,000
Molecular weight: ~ 496
Excitation source: UV
Replaces: Alexa Fluor® 350, AMCA, DyLight® 350

Figure 1. Absorption and emission spectra of CF350 goat anti-mouse conjugate in PBS.



Features

- Brighter and more photostable than AMCA
- Direct replacement for Alexa Fluor® 350
- Highly water soluble and pH-insensitive

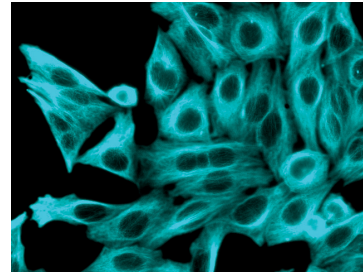


Figure 2. HeLa cells stained with mouse anti-tubulin antibody and CF350 goat anti-mouse IgG (cyan).

CF™ 405S and CF™ 405M

Improved brightness and photostability for the 405 nm laser line

Technical Summary

CF™ 405S

Abs/Em Maxima: 404/431 nm
Extinction coefficient: 33,000
Molecular weight: ~ 1,169
Excitation laser line: 405 nm
Replaces: Alexa Fluor® 405, Cascade Blue®, DyLight® 405

CF™ 405M

Abs/Em Maxima: 408/452 nm
Extinction coefficient: 41,000
Molecular weight: ~ 503
Excitation laser line: 405 nm
Replaces: Pacific Blue®, BD Horizon™ V450

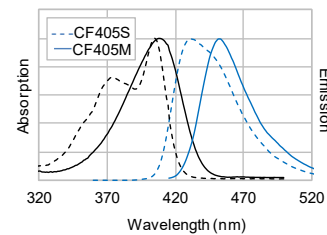


Figure 1. Absorption and emission spectra of CF405S and CF405M goat anti-mouse conjugates in PBS.

Features

- CF™ 405S: Brighter than Alexa Fluor® 405
- CF™ 405M: More photostable than Pacific Blue®, with less spill-over in the green channel
- CF™ 405M: an excellent choice for super-resolution imaging by SIM (see pp. 20-21)

Figure 2. Intracellular staining of Jurkat cells was performed with mouse anti-CD3 or isotype control followed by goat anti-mouse IgG conjugated to Alexa Fluor 405 (AF405) or CF405S. Fluorescence was analyzed on a BD LSR II flow cytometer with 405 nm excitation and 450/50 nm emission filter. Bars represent the relative fluorescence of the geometric means of the cell populations.

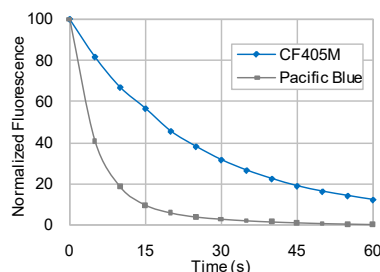
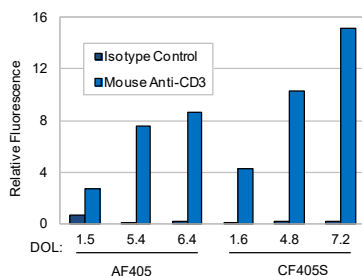


Figure 3. Photostability of CF405M and Pacific Blue. CF405M and Pacific Blue dye solutions were continuously exposed to mercury arc lamp microscope excitation with a DAPI filter set. Images were captured every 5 seconds for one minute. Fluorescence intensity was normalized to time 0.

CF™ 405L Dye

A 405 nm-excitable dye with orange fluorescence emission

Technical Summary

Abs/Em Maxima: 395/545 nm
 Extinction coefficient: 24,000
 Molecular weight: ~1573
 Excitation laser line: 405 nm
 Replaces: Pacific Orange®

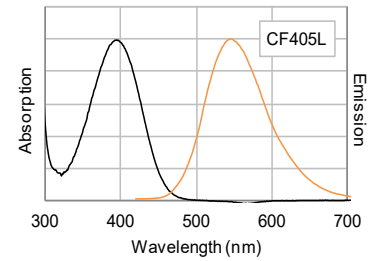


Figure 1. Absorption and emission spectra of CF405L goat anti-mouse conjugate in PBS.

CF™ 430 and CF™ 440

Photostable 405 nm-excitable dyes with green fluorescence

Technical Summary

CF™ 430

Abs/Em Maxima: 426/498 nm
 Extinction coefficient: 40,000
 Molecular weight: ~429
 Excitation laser line: 405 nm
 Replaces: Pacific Green®, BD Horizon™ V500, Krome Orange™

CF™ 440

Abs/Em Maxima: 440/515 nm
 Extinction coefficient: 40,000
 Molecular weight: ~716
 Excitation laser line: 405 nm
 Replaces: Alexa Fluor® 430

Features

- Photostable dyes suitable for microscopy
- CF430 is a perfect match for the CFP filter set
- Suitable for flow cytometry in the AmCyan channel
- Highly water soluble and pH-insensitive

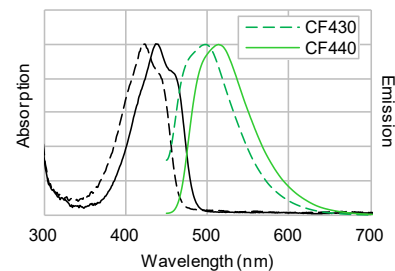


Figure 1. Absorption and emission spectra of CF430 and CF440 goat anti-mouse conjugates in PBS.

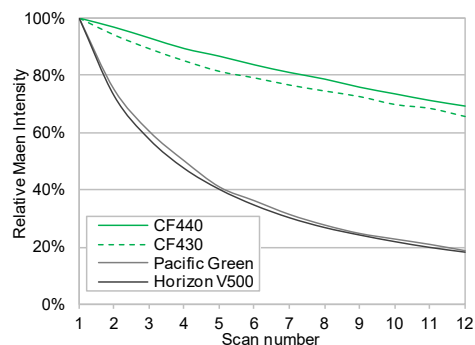


Figure 2. Relative photostability of CF430 and CF440 compared to spectrally-similar dyes. Cells were stained with biotinylated primary antibodies followed by streptavidin conjugates of CF430, CF440, Pacific Green, or BD Horizon V500. Fluorescence was imaged on a Zeiss LSM700 confocal microscope in the FITC channel using 405 nm excitation. Images were acquired every 5 seconds for 12 consecutive scans of the same field of view using the same imaging settings for each dye. The mean fluorescence intensity of each image was normalized to the first scan for each dye.

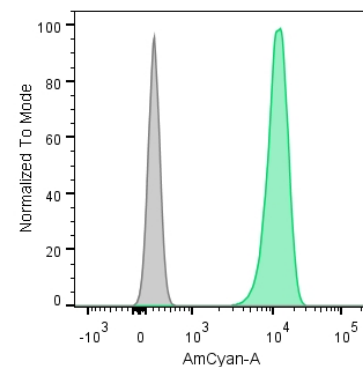


Figure 3. Flow cytometry analysis of Jurkat cells stained with isotype control (gray peak) or mouse anti-CD3 (green peak) followed by CF430 goat anti-mouse IgG, analyzed in the AmCyan channel of a BD LSRII flow cytometer.

CF™450

A green fluorescent dye with unique spectral properties

Technical Summary

Abs/Em Maxima: 450/538 nm

Extinction coefficient: 40,000

Molecular weight: ~689

Excitation laser line: 405 nm

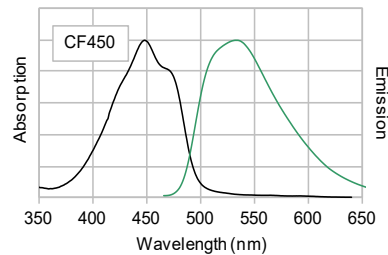


Figure 1. Absorption and emission of CF450 goat anti-mouse conjugate in PBS.

CF™488A

A superior green fluorescent dye

Technical Summary

Abs/Em Maxima: 490/515 nm

Extinction coefficient: 70,000

Molecular weight: ~914

Excitation laser line: 488 nm

Replaces: Alexa Fluor® 488, DyLight® 488, fluorescein (aka FITC, FAM), Cy@2

Features

- Minimally charged, for less non-specific binding than Alexa Fluor® 488
- Narrower emission spectrum for less bleed to red
- Very photostable
- Compatible with super-resolution imaging by TIRF (see pp. 20-21)
- Highly water soluble and pH-insensitive

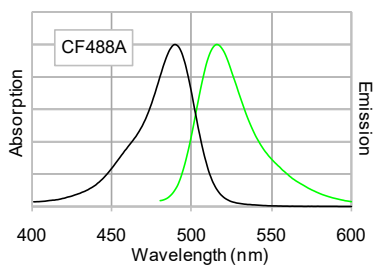


Figure 1. Absorption and emission spectra of CF488A goat anti-mouse conjugate in PBS.

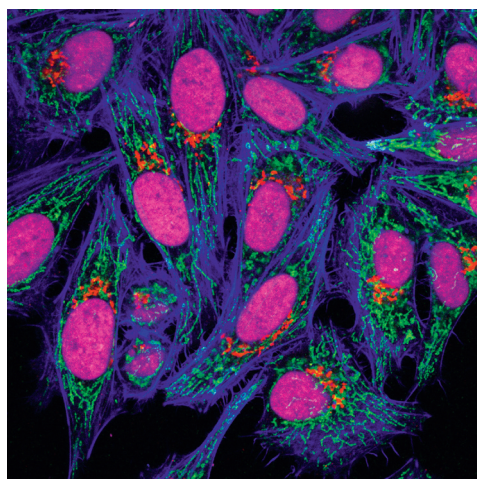


Figure 2. HeLa cells stained with rabbit anti-COXIV and CF488A goat anti-rabbit IgG (mitochondria, green), mouse anti-Golgin 97 and CF™555 goat anti-mouse IgG (Golgi, red), CF405M phalloidin (actin filaments, blue), and RedDot2 (nuclei, magenta). See p. 30 for more information on RedDot2.

CFTM514

Alternative green fluorescent dye

Technical Summary

Abs/Em Maxima: 516/548 nm

Extinction coefficient: 105,000

Molecular weight: ~1216

Excitation laser line: 488 nm

Replaces: Alexa Fluor® 514

Features

- Image using the same settings as FITC or CFTM488A
- Can be distinguished from CFTM488A in the same specimen by spectral imaging and linear unmixing

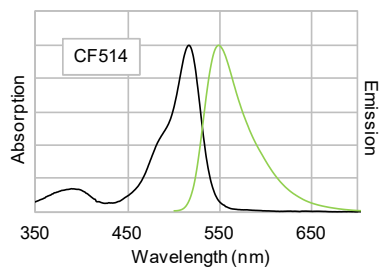


Figure 1. Absorption and emission spectra of CF514 goat anti-mouse conjugate in PBS.

CF™ 532

A bright orange fluorescent dye for the 532 nm laser

Technical Summary

Abs/Em Maxima: 527/558 nm
 Extinction coefficient: 96,000
 Molecular weight: ~685
 Excitation laser line: 532 nm
 Direct replacement for: Alexa Fluor® 532, Atto 532

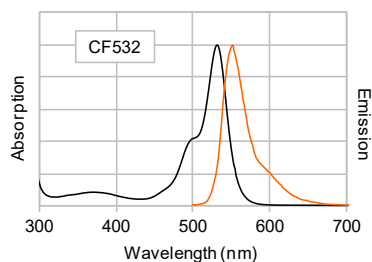


Figure 1. Absorption and emission spectra of CF532 goat anti-mouse IgG conjugate in PBS.

Features

- Designed for the 532 nm laser
- Brighter than Alexa Fluor® 532 (Fig. 2)
- Highly water-soluble and pH-insensitive

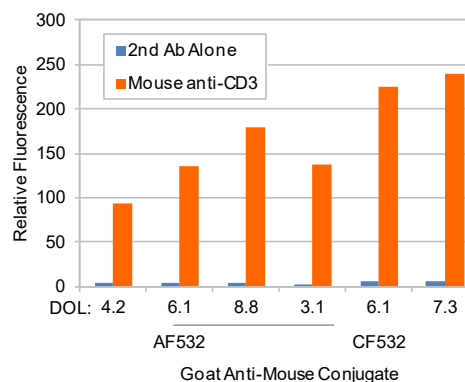


Figure 2. Flow cytometry analysis of Jurkat cells stained with Alexa Fluor 532 (AF532) antibody or CF532 secondary antibody conjugates. Intracellular staining was performed with mouse anti-CD3 antibody followed by goat anti-mouse IgG conjugates. Background was determined by staining with secondary antibody (2nd Ab) alone. Fluorescence was analyzed on a BD FACSCalibur flow cytometer in the FL2 channel. The bars represent the relative fluorescence of the geometric means of the cell populations.

CF™ 535ST

An orange fluorescent dye designed for STORM super-resolution imaging

Technical Summary

Abs/Em Maxima: 535/568 nm
 Extinction coefficient: 95,000
 Molecular weight: ~728
 Excitation laser line: 532 nm

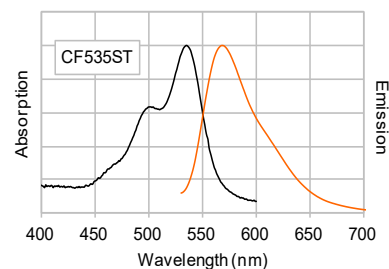


Figure 1. Absorption and emission spectra of CF535ST goat anti-mouse IgG conjugate in PBS.

See pp. 20-21 for more information about CF™ dyes for super-resolution imaging.

CF™ 543

An orange fluorescent dye ideal for the 543 nm laser

Technical Summary

Abs/Em Maxima: 541/560 nm
 Extinction coefficient: 100,000
 Molecular weight: ~ 870
 Excitation laser line: 532 nm, 543 nm, or 546 nm
 Direct replacement for: Alexa Fluor® 546, TAMRA

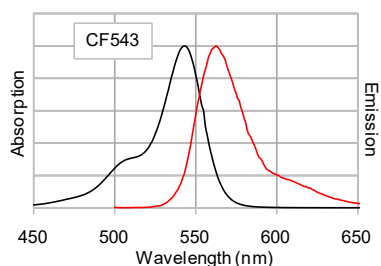


Figure 1. Absorption and emission spectra of CF543 goat anti-mouse conjugate in PBS.

Features

- Optimized for the 543 nm laser
- Yields the brightest conjugates among spectrally similar dyes
- Highly water-soluble and pH-insensitive

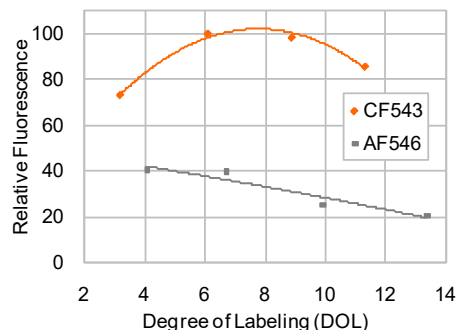


Figure 2. Relative fluorescence of CF543 and Alexa Fluor 546 (AF546) goat anti-mouse conjugates as a function of the number of dye molecules per protein (degree of labeling).

CF™ 555

A bright and photostable orange-red dye

Technical Summary

Abs/Em Maxima: 555/565 nm
 Extinction coefficient: 150,000
 Molecular weight: ~ 901
 Excitation laser line: 532 nm or 568 nm
 Direct replacement for: Alexa Fluor®555, ATTO 550, Cy@3, DyLight® 549, Rhodamine

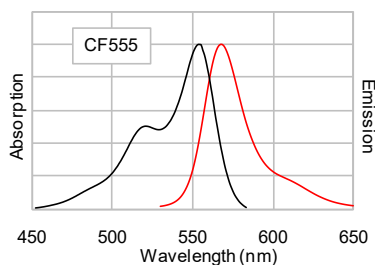


Figure 1. Absorption and emission spectra of CF555 goat anti-mouse conjugate in PBS.

Features

- Brighter than Cy@3
- Highly water-soluble
- Validated in multicolor STORM super-resolution imaging (see pp. 20-21)

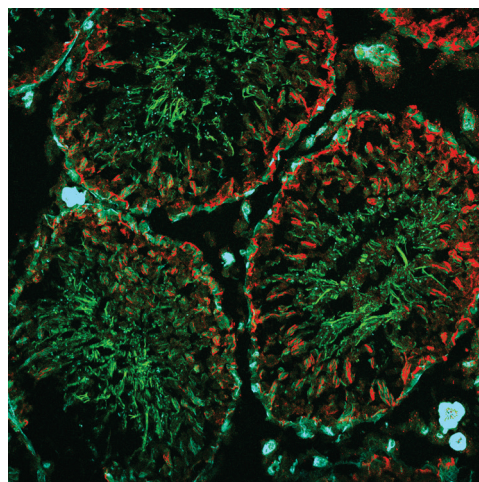


Figure 2. Frozen section of rat testis stained with mouse anti-tubulin and CF488A goat anti-mouse (min x rat) (microtubules, green), CF555 Mix-n-Stain labeled mouse anti-ZO1 (tight junctions, red) and CF640R phalloidin (actin filaments, cyan). See p. 23 for more information on Mix-n-Stain antibody labeling kits.

CF™568

Outshines Alexa Fluor®568

Technical Summary

Abs/Em Maxima: 562/583 nm
 Extinction coefficient: 100,000
 Molecular weight: ~ 714
 Excitation laser line: 532 nm or 568 nm
 Direct replacement for: Alexa Fluor® 568, ATTO 565, Rhodamine Red

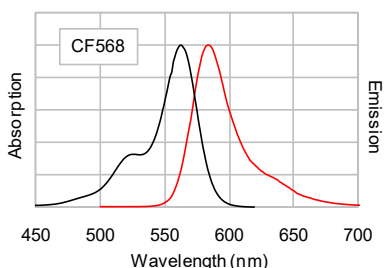


Figure 1. Absorption and emission spectra of CF568 goat anti-mouse conjugate in PBS.

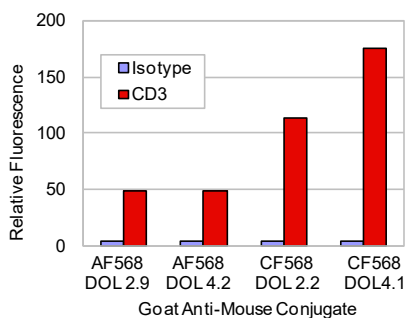


Figure 2. Intracellular staining of Jurkat cells was performed using mouse anti-CD3 or isotype control followed by goat anti-mouse IgG conjugates. Fluorescence was analyzed on a BD FACSCalibur flow cytometer in the FL2 channel. Bars represent the relative fluorescence of the geometric means of the cell populations.

Features

- Yields much brighter antibody conjugates than Alexa Fluor® 568
- Extremely photostable
- Excellent choice for multi-color imaging with CF™488A and CF™640R
- Compatible with TIRF and multicolor STORM super-resolution imaging (see pp. 20-21)

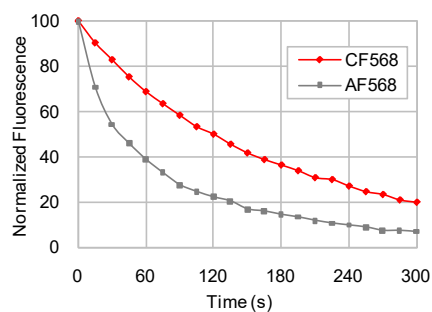


Figure 3. Photostability of CF568 and Alexa Fluor 568 (AF568) streptavidin conjugates. Intracellular staining of Jurkat cells was performed using anti-CD3-biotin followed by streptavidin-CF568 or streptavidin-AF568. Cells were continuously exposed to mercury arc lamp microscope excitation with a Cy3 filter set. Images were captured every 15 seconds for 5 minutes and fluorescence intensity was normalized to time 0.

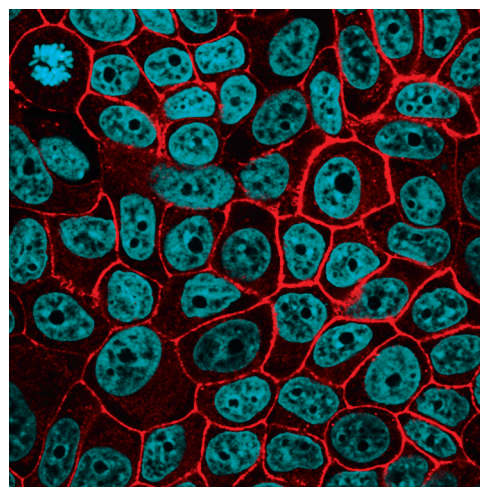


Figure 4. MCF-7 cells stained with CF568 mono-clonal anti-Ep-CAM (clone EGP40/826) at 5 ug/mL (red). Nuclei are counterstained with Hoechst 33342 (blue). See p. 26 for more information on primary antibody conjugates.

CF™594

Truly the brightest deep red dye

Technical Summary

Abs/Em Maxima: 593/614 nm

Extinction coefficient: 115,000

Molecular weight: ~730

Excitation laser line: 532 nm, 568 nm or 594 nm

Replaces: Alexa Fluor® 594, DyLight® 594, Texas Red®

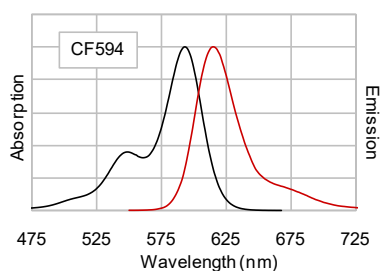


Figure 1. Absorption and emission spectra of CF594 goat anti-mouse conjugate in PBS.

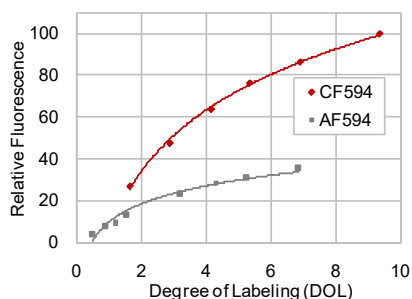


Figure 2. Relative fluorescence of CF594 and Alexa Fluor 594 (AF594) goat anti-mouse conjugates as a function of the number of dye molecules per protein (degree of labeling).

Features

- Yields the brightest antibody conjugates among spectrally similar dyes.
- Excellent choice for multicolor imaging with green dyes like CF™488A
- Extremely photostable
- Also see CF™594ST, a version of CF™594 engineered specifically for STORM microscopy (pp. 20-21).

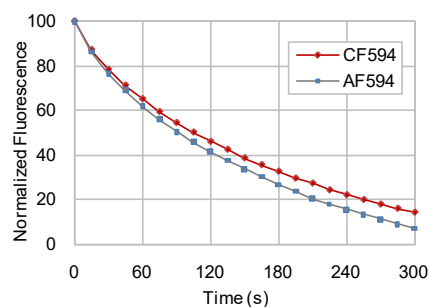


Figure 3. Photostability of CF594 and Alexa Fluor 594 (AF594) goat anti-mouse conjugates. Intracellular staining of Jurkat cells was performed with mouse anti-CD3 followed by CF594 or AF594 goat anti-mouse conjugates. Cells were continuously exposed to mercury arc lamp microscope excitation with a Texas Red filter set. Images were captured every 15 seconds for 5 min and fluorescence intensity was normalized to time 0.

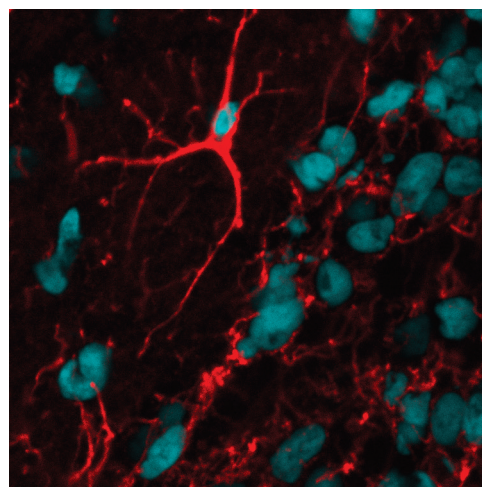


Figure 4. Glial cells in frozen section of rat brain stained with rabbit anti-GFAP antibody and CF594 goat anti-rabbit IgG (red). Nuclei are stained with RedDot2 (pseudocolored cyan). Mounted with Everbrite Mounting Medium. See p. 30 for more information on RedDot2 and EverBrite Mounting Medium.

CF™620R

A bright and photostable far-red dye

Technical Summary

Abs/Em Maxima: 617/639 nm

Extinction coefficient: 115,000

Molecular weight: ~ 738

Excitation laser line: 633 nm or 635 nm

Replaces: LightCycler® Red 640

Features

- Highly water-soluble
- Highly fluorescent and extremely photostable
- Absorption/emission at 617/639 nm for use in FRET or multi-color detection

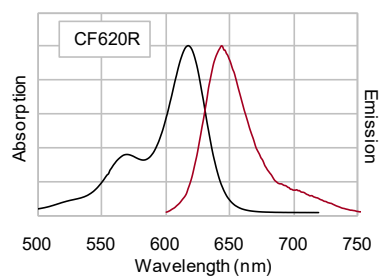


Figure 1. Absorption and emission spectra of CF620R free acid in PBS.

CF™ 633

The best dye for 633/635 laser lines

Technical Summary

Abs/Em Maxima: 630/650 nm

Extinction coefficient: 100,000

Molecular weight: ~820

Excitation laser line: 633 nm or 635 nm

Replaces: Alexa Fluor® 633, Alexa Fluor® 647, Cy@5, DyLight® 633, DyLight® 649

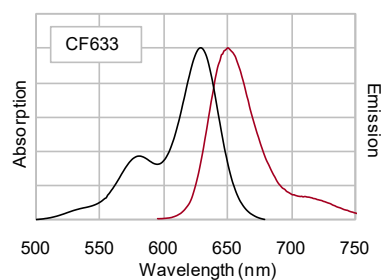


Figure 1. Absorption and emission spectra of CF633 goat anti-mouse conjugate in PBS.

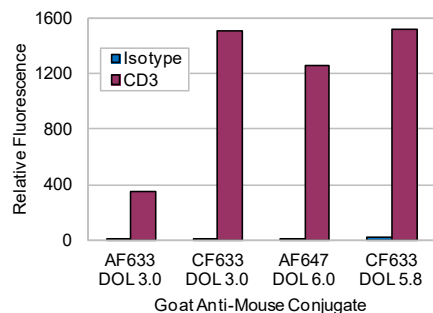


Figure 2. Intracellular staining of Jurkat cells was performed using mouse anti-CD3 or isotype control followed by goat anti-mouse IgG conjugates. Fluorescence was analyzed on a BD FACSCalibur flow cytometer in the FL4 channel. Bars represent the relative fluorescence of the geometric means of the cell populations.

Features

- Yields the brightest antibody conjugates among spectrally similar dyes
- Far more photostable than Alexa Fluor® 647
- Compatible with TIRF, FIONA, and gSHRIMP super-resolution imaging methods (see pp. 20-21)

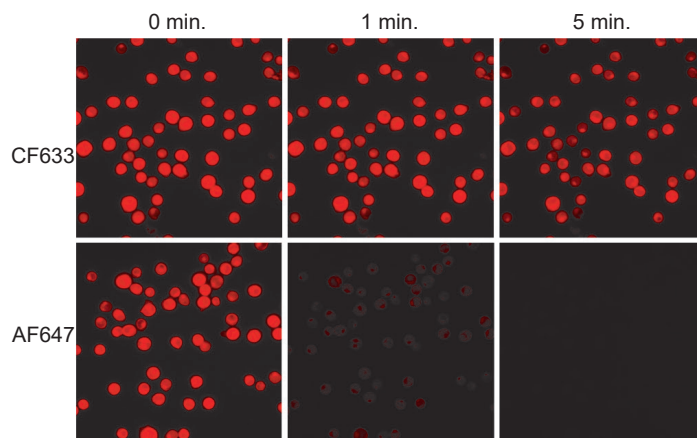


Figure 3. Relative photostability of CF633 and Alexa Fluor 647 (AF647) goat anti-mouse conjugates. Jurkat cells were fixed, permeabilized and stained with rabbit anti-CD3 followed by CF633 or Alexa Fluor 647 goat anti-rabbit IgG conjugates. Cells were imaged using a mercury arc lamp microscope equipped with a Cy5 filter set and CCD camera. Sequential images were captured at 0, 1, and 5 minutes.

CF™640R

A highly photostable far-red dye

Technical Summary

Abs/Em Maxima: 642/662 nm
 Extinction coefficient: 105,000
 Molecular weight: ~ 832
 Excitation laser line: 633 nm, 635 nm or 640 nm
 Replaces Alexa Fluor® 647, ATTO 647N, Cy®5, DyLight® 649

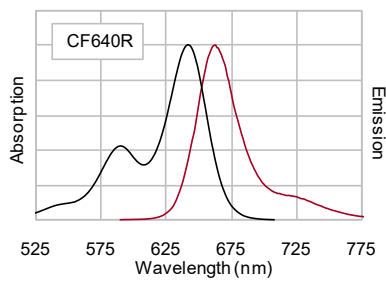


Figure 1. Absorption and emission spectra of CF640R goat anti-mouse conjugate in PBS.

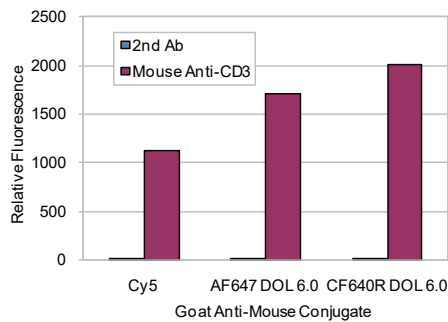


Figure 2. Intracellular staining of Jurkat cells was performed using mouse anti-CD3 or isotype control followed by goat anti-mouse IgG conjugates. Fluorescence was analyzed on a BD FACSCalibur flow cytometer in the FL4 channel. Bars represent the relative fluorescence of the geometric means of the cell populations.

Features

- Best photostability among Cy®5-like dyes
- Yields highly fluorescent protein conjugates
- Compatible with TIRF and FLIMP super-resolution microscopy (see pp. 20-21)

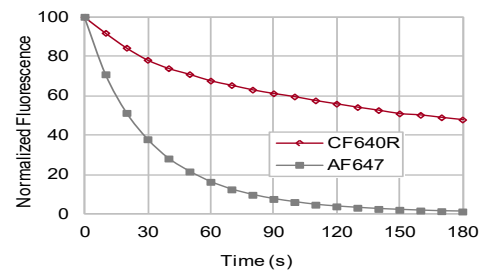


Figure 3. Photostability comparison between CF640R and Alexa Fluor 647 (AF647). HeLa cells were stained with anti-tubulin antibody conjugates of each dye. Cells were continuously illuminated by a mercury arc lamp microscope and sequential images were captured at 0, 1, and 3 minutes. Mean fluorescence was normalized to time 0.

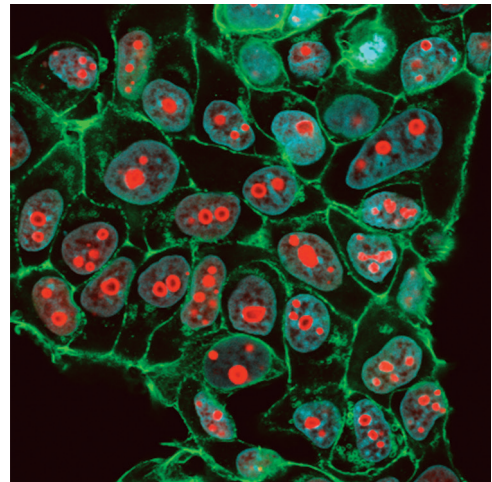


Figure 4. MCF-7 cells stained with CF640R monoclonal anti-Cyclin B1 (clone CCNB1/1098) at 5 µg/mL (red). Nuclei are counterstained with Hoechst 33342 (blue) and actin filaments are stained with CF488A phalloidin (green). See p. 26 for more information on primary antibody conjugates.

CF™647

A highly fluorescent far-red dye

Technical Summary

Abs/Em Maxima: 650/665 nm

Extinction coefficient: 240,000

Molecular weight: ~ 1058

Excitation laser line: 633 nm, 635 nm or 640 nm

Replaces: Cy®5, Alexa Fluor® 647, DyLight® 649

Features

- Brighter than Cy®5
- Highly water soluble and pH insensitive
- Validated in multi-color super-resolution imaging by STORM (see pp. 20-21)

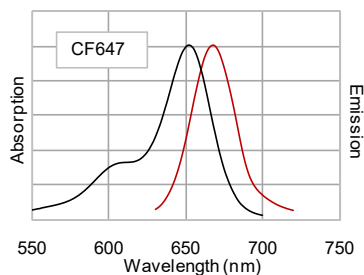


Figure 1. Absorption and emission spectra of CF647 goat anti-mouse conjugate in PBS.

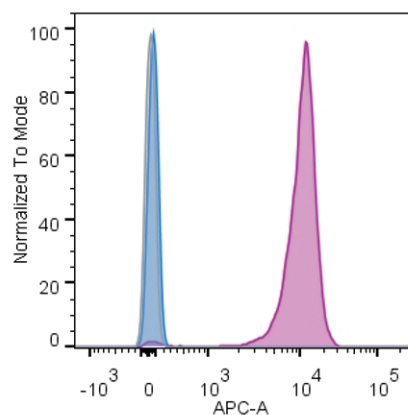


Figure 2. Intracellular staining of Jurkat cells with CF647 monoclonal anti-nucleolin (clone 365-2) (pink) or CF647 IgG1 isotype control (blue) at 1 ug/tube, compared to unstained cells (yellow). Cells were analyzed in the APC channel of a BD LSRII flow cytometer. See p. 26 for more information on primary antibody conjugates.

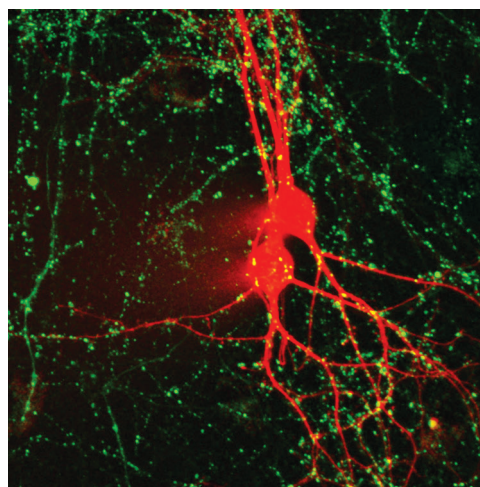


Figure 3. Cultured rat hippocampal neurons microinjected with CF647 hydrazide (red) and stained with SynaptoGreen™ C4 (FM1-43) (green, synaptic vesicles). Image courtesy of Professor Guosong Liu, Tsinghua University, Beijing, China.

CF™660C and CF™660R

Superior alternatives to Alexa Fluor® 660

Technical Summary

CF™660C

Abs/Em Maxima: 667/685 nm
 Extinction coefficient: 200,000
 Molecular weight: ~ 3112
 Excitation laser line: 633 nm, 635 nm or 640 nm
 Replaces: Alexa Fluor® 660, Allophycocyanin (APC)

CF™660R

Abs/Em Maxima: 663/682 nm
 Extinction coefficient: 100,000
 Molecular weight: ~ 888
 Excitation laser line: 633 nm, 635 nm or 640 nm
 Replaces: Alexa Fluor® 660, Allophycocyanin (APC)

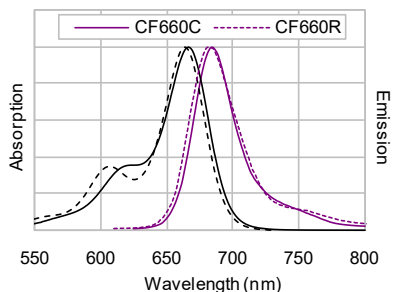


Figure 1. Absorption and emission spectra of CF660C and CF660R goat anti-mouse conjugates in PBS.

CF™660C Features

- Much brighter and more photostable than Alexa Fluor® 660
- Compatible with multicolor super-resolution imaging by STORM (see pp. 20-21)

CF™660R Features

- Brighter than Alexa Fluor® 660
- Unrivaled photostability among spectrally similar dyes

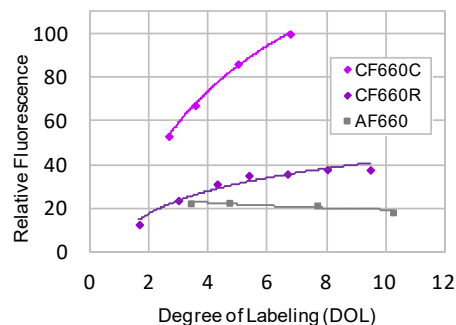


Figure 2. Relative fluorescence of CF660, CF660R and Alexa Fluor 660 (AF660) goat anti-mouse conjugates as a function of the number of dye molecules per protein (degree of labeling).

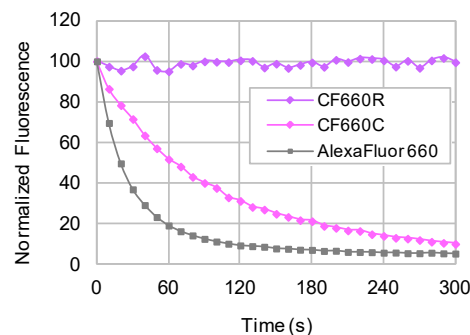


Figure 3. Photostability of CF660C, CF660R, and Alexa Fluor 660 (AF660) goat anti-mouse conjugates. HeLa cells were stained with mouse anti-tubulin followed by CF660C, CF660R or AF660 goat anti-mouse IgG conjugates. Cells were continuously exposed to mercury arc lamp microscope excitation using a Cy5 filter set. Images were captured every 10 seconds for five minutes and fluorescence intensity was normalized to time 0.

CF™ 680 and CF™ 680R Dyes

Two outstanding 680 nm-excitable dyes

Technical Summary

CF™ 680

Abs/Em Maxima: 681/698 nm

Extinction coefficient: 210,000

Molecular weight: ~ 3241

Excitation laser line: 680 nm or 685 nm

Replaces: Alexa Fluor® 680, Cy®5.5, IR®Dye 680

CF™ 680R

Abs/Em Maxima: 680/701 nm

Extinction coefficient: 140,000

Molecular weight: ~ 912

Excitation laser line: 680 nm or 685 nm

Replaces: Alexa Fluor® 680, Cy®5.5, IR®Dye 680

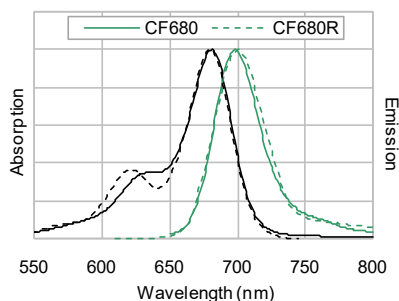


Figure 1. Absorption and emission spectra of CF680 and CF680R goat anti-mouse conjugates in PBS.

CF™ 680 Features

- The brightest among spectrally similar dyes
- Validated in multicolor STORM and 3D super-resolution microscopy (see pp. 20-21)
- Compatible with LI-COR® Odyssey®

CF™ 680R Features

- Unrivaled photostability among spectrally similar dyes
- Compatible with STED and single molecule spectroscopy super-resolution imaging (see pp. 20-21)
- Molecular weight compatible with nucleic acid labeling
- Compatible with LI-COR® Odyssey®

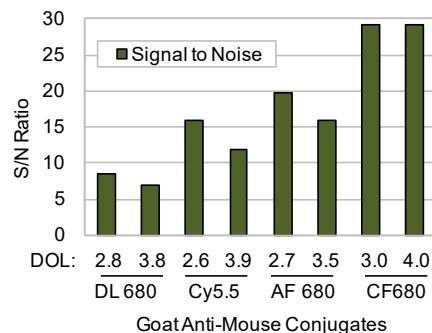


Figure 2. Intracellular staining of Jurkat cells was performed using mouse anti-human CD3 antibody or isotype control followed by goat anti-mouse secondary antibody conjugates. Fluorescence was analyzed on a BD FACSCalibur flow cytometer in the FL4 channel. Bars represent the signal-to-noise ratio of CD3-positive fluorescence to isotype control.

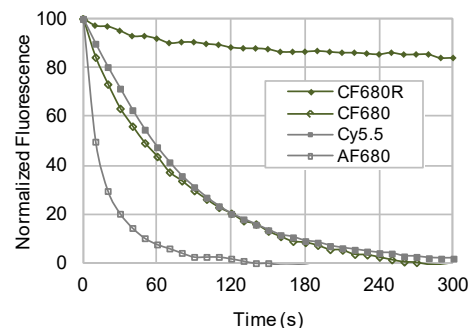


Figure 3. Photostability of far red dye conjugates. Jurkat cells were stained with mouse anti-CD3 followed by the indicated goat anti-mouse IgG conjugates. Cells were continuously exposed to mercury arc lamp excitation with a Cy5 filter set. Images were captured every 10 seconds for 5 minutes and fluorescence intensity was normalized to time 0.

CF™ 750, CF™ 770, CF™ 790, and CF™ 800

Unrivaled Near-Infrared Dyes

Technical Summary

CF™ 750

Abs/Em Maxima: 755/777 nm
 Extinction coefficient: 250,000
 Molecular weight: ~ 3009
 Excitation laser line: 633 nm, 635 nm, 680 nm or 685 nm
 Replaces: Alexa Fluor® 750, Cy®7, DyLight® 750

CF™ 770

Abs/Em Maxima: 770/797 nm
 Extinction coefficient: 220,000
 Molecular weight: ~ 3138
 Excitation laser line: 785 nm
 Replaces: DyLight™ 800, IRDye 800CW

CF™ 790

Abs/Em Maxima: 784/806 nm
 Extinction coefficient: 210,000
 Molecular weight: ~ 3267
 Excitation laser line: 785 nm
 Replaces: Alexa Fluor® 790

CF™ 800

Abs/Em Maxima: 797/816 nm
 Extinction coefficient: 210,000
 Molecular weight: ~3334
 Excitation laser line: 785 nm
 Spectrally similar to: Indocyanine Green

Features

- Exceptionally bright and stable
- Ideal for in vivo imaging
- Compatible with LI-COR® Odyssey®
- Superior signal-to-noise for bioconjugates
- CF™ 750 validated in STORM microscopy (see pp. 20-21)

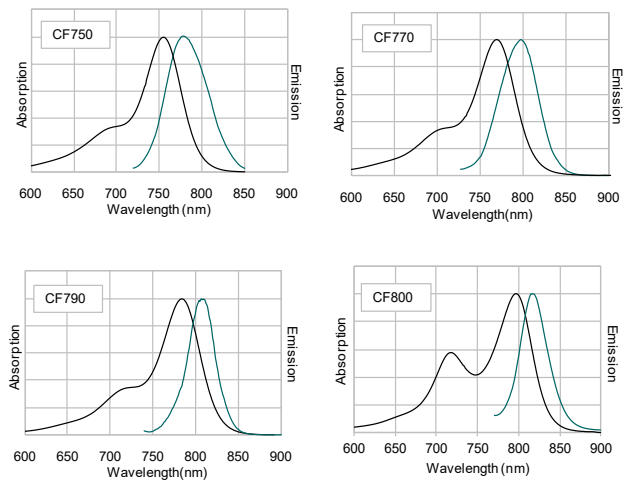


Figure 1. Absorption and emission spectra of near-IR CF dye goat anti-mouse conjugates in PBS.

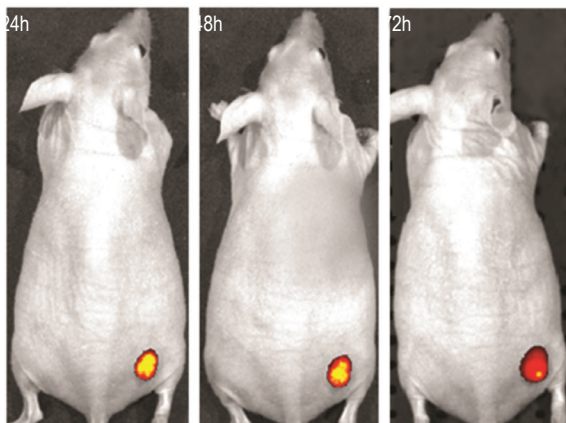


Figure 2. Tumors in mice were imaged using an IVIS® imaging system (Caliper Life Sciences) 24 hours, 48 hours, and 96 hours after IV injection of Avastin conjugated to CF750. Images courtesy of Caliper Life Sciences.

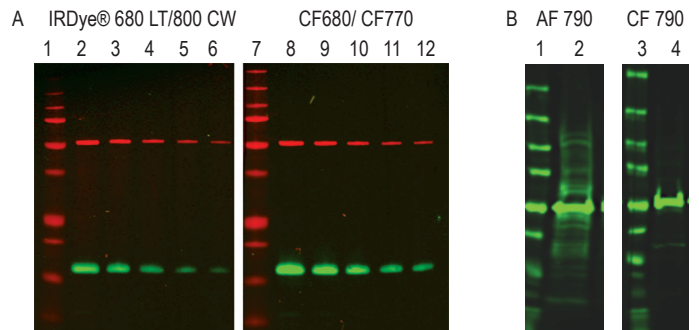


Figure 3. Near-infrared western blotting with CF dyes compared to spectrally similar dyes. A. Two-fold dilutions of HeLa cell lysate containing 2 ug, 1 ug, 0.5 ug, 0.25 ug, 0.125 ug total protein per lane were separated by SDS-PAGE, transferred to Immobilon FL PVDF (Millipore), and probed with mouse anti-tubulin and rabbit anti-COXIV antibodies. Secondary detection was performed with either IRDye® 680LT goat anti-mouse (red) and IRDye® 800CW goat anti-rabbit (green) (LI-COR; lanes 1-6) or CF680 goat anti-mouse (red) and CF770 goat anti-rabbit (green) (lanes 7-12) at the same final concentrations. Membranes were scanned using an Odyssey® infrared imaging system. Quantitation of the bands showed approximately 1.5-2-fold higher fluorescence intensity of CF dye secondary antibodies compared to IRDye secondary antibodies. Lanes 1 and 7 contain Odyssey Molecular Weight Marker (LI-COR Biosciences). B. Western blots of HeLa cell lysate (lanes 2 and 4) were probed with mouse anti-tubulin antibody followed by goat anti-mouse conjugated to Alexa Fluor 790 (AF790, left) or CF790 (right). CF790 does not introduce excessive negative charge to antibody conjugates, which can increase non-specific binding. Lanes 1 and 3 contain DyLight 680/800 Protein Ladder (Pierce).

IRDye, LI-COR, Odyssey, and In-Cell Western are trademarks or registered trademarks of LI-COR, Inc. in the United States and other countries. IVIS is a registered trademark of Caliper Life Sciences.

CF™ Dyes for Super-Resolution Imaging

Recent publications comparing synthetic dyes for super-resolution imaging have shown 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 and single molecule imaging techniques. Biotium's CF™405M has been found to be the brightest and most photostable short wavelength fluorescent dye for SIM. Six 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 for CF™ dye single-molecule imaging applications on page 21. Biotium offers a wide selection of CF™ dye labeled secondary antibodies, other conjugates, and labeling kits; visit www.biotium.com for our full selection of products.

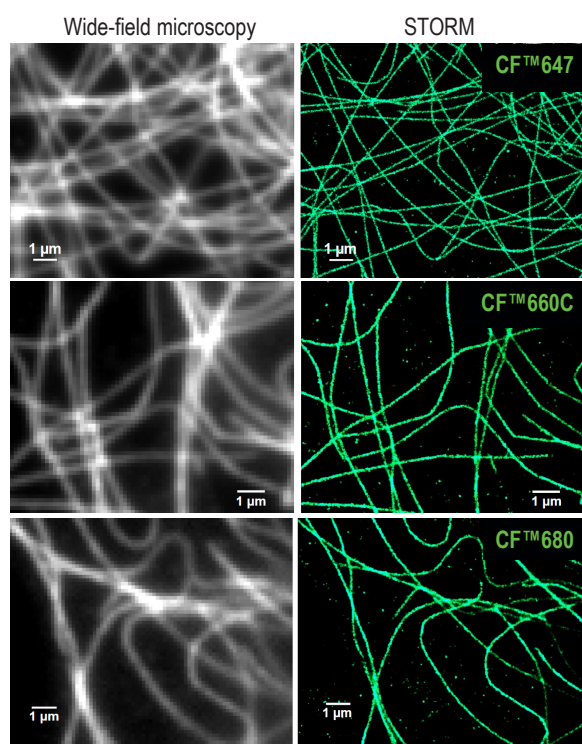


Figure 1. Comparison of conventional wide-field microscopy (left) with STORM (right) using CF™ dye conjugates. Fixed cells were stained with mouse anti-tubulin antibody followed by CF™ dye conjugated anti-mouse secondary antibody (top row: CF™647, middle row: CF™660C, bottom row: CF™680). For STORM, 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.

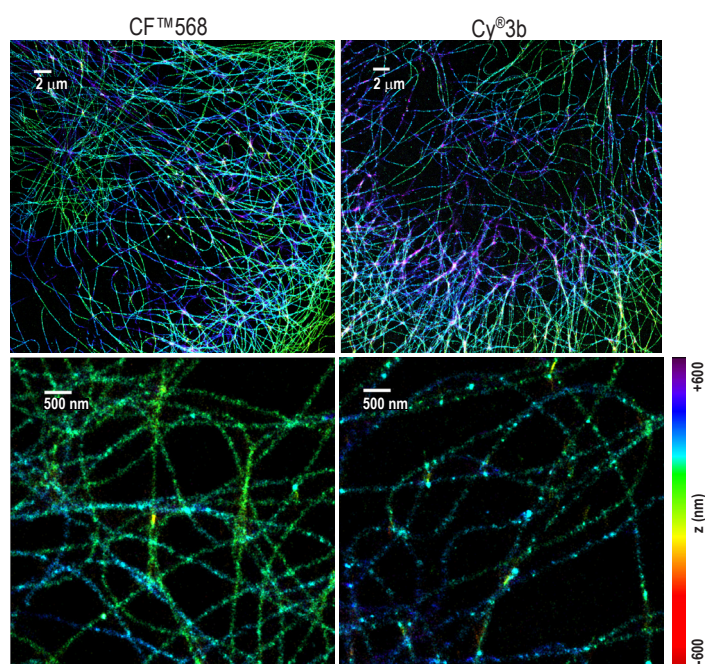


Figure 2. CF™568 (left) produces better images than Cy®3b (right) in 3-D STORM microscopy. Fixed cells were stained with mouse anti-tubulin antibody followed by dye-conjugated anti-mouse secondary antibodies. See Figure 1 legend for imaging conditions. Dye molecules were photoswitched and imaged using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

CF™ Dyes for Super-Resolution Imaging

Super-resolution imaging techniques validated for CF™ dyes

CF™ Dye	Abs/Em maxima (nm)	Extinction coefficient	Super resolution application	References
CF™405M	408/452	41,000	SIM	Markaki, Y. et al. (2013). Fluorescence In Situ Hybridization Applications for Super-Resolution 3D Structured Illumination Microscopy . <i>Methods Mol Biol</i> 950, 43-64.
CF™488A	490/515	70,000	TIRF	Zanetti-Domingues, L.C. et al. (2013). Hydrophobic Fluorescent Probes Introduce Artifacts into Single Molecule Tracking Experiments Due to Non-Specific Binding . <i>PLoS ONE</i> 8(9): e74200.
CF™535ST	535/568	95,000	STORM	Collaborator communication; contact techsupport@biotium.com for more information.
CF™555	555/565	150,000	Multicolor STORM	Lehmann, M. et al. (2015). Novel organic dyes for multicolor localization-based super-resolution microscopy . <i>J Biophotonics</i> DOI 10.1002/jbio.201500119
CF™568	562/583	100,000	TIRF, multicolor STORM	Lehmann, M. et al. (2015). Novel organic dyes for multicolor localization-based super-resolution microscopy . <i>J Biophotonics</i> DOI 10.1002/jbio.201500119 Zanetti-Domingues, L.C. et al. (2013). Hydrophobic Fluorescent Probes Introduce Artifacts into Single Molecule Tracking Experiments Due to Non-Specific Binding . <i>PLoS ONE</i> 8(9): e74200. Zhang, M. et al. (2015). Translocation of interleukin-1β into a vesicle intermediate in autophagy-mediated secretion . <i>eLife</i> 2015;10.7554/eLife.11205
CF™594ST	593/614	115,000	STORM	Collaborator communication; contact techsupport@biotium.com for more information.
CF™633	630/650	100,000	TIRF, FIONA, gSHRImP	Bosch, P. J. et al. (2014). Evaluation of fluorophores to label SNAP-tag fused proteins for multicolor single-molecule tracking microscopy in live cells . <i>Biophys J</i> 107, 803-814. Zanetti-Domingues, L.C. et al. (2013). Hydrophobic Fluorescent Probes Introduce Artifacts into Single Molecule Tracking Experiments Due to Non-Specific Binding . <i>PLoS ONE</i> 8(9): e74200. Kim, H. J., and Selvin, P. R. (2013). Fluorescence Imaging with One Nanometer Accuracy . SpringerReference Encyclopedia of Biophysics Simonson, P. D., Rothenberg, E., and Selvin, P. R. (2011). Single-molecule-based super-resolution images in the presence of multiple fluorophores . <i>Nano Lett</i> 11, 5090-5096. DOI:10.1021/nl203560r
CF™640R	642/662	105,000	TIRF, FLImP	Bosch, P. J. et al. (2014). Evaluation of fluorophores to label SNAP-tag fused proteins for multicolor single-molecule tracking microscopy in live cells . <i>Biophys J</i> 107, 803-814. Martin-Fernandez, M. L. et al. (2013). A 'pocket guide' to total internal reflection fluorescence . <i>J Microsc</i> 252, 16-22. Zanetti-Domingues, L.C. et al. (2013). Hydrophobic Fluorescent Probes Introduce Artifacts into Single Molecule Tracking Experiments Due to Non-Specific Binding . <i>PLoS ONE</i> 8(9): e74200. Needham, S.R., et al. (2015). Determining the geometry of oligomers of the human epidermal growth factor family on cells with <10 nm resolution . <i>Biochem Soc Trans</i> 43, 309–314.
CF™647	650/665	240,000	Multicolor STORM	Lehmann, M. et al. (2015). Novel organic dyes for multicolor localization-based super-resolution microscopy . <i>J Biophotonics</i> DOI 10.1002/jbio.201500119 Olivier, N. et al. (2013). Simple buffers for 3D STORM microscopy . <i>Biomed Opt Express</i> 4, 885-899.
CF™660C	667/685	200,000	Multicolor STORM	Zhang, Z., et al. (2015). Ultrahigh-throughput single-molecule spectroscopy and spectrally resolved super-resolution microscopy . <i>Nature Methods</i> doi:10.1038/nmeth.3528
CF™680	681/698	210,000	Multicolor STORM, dual-color 3D super resolution microscopy	Früh, S.M. et al. (2015). Molecular architecture of native fibronectin fibrils . <i>Nature Communications</i> 6, 7275. Lehmann, M. et al. (2015). Novel organic dyes for multicolor localization-based super-resolution microscopy . <i>J Biophotonics</i> DOI 10.1002/jbio.201500119 Platonova, E. et al. (2015). A Simple Method for GFP- and RFP-based Dual Color Single-Molecule Localization Microscopy . <i>ACS Chem. Biol.</i> 10(6),1411–1416. Platonova, E. et al. (2015). Single-molecule microscopy of molecules tagged with GFP or mRFP derivatives in mammalian cells using nanobody binders . <i>Methods</i> doi: http://dx.doi.org/10.1016/j.ymeth.2015.06.018 Salvador-Gallego, R. et al. (2016). Bax assembly into rings and arcs in apoptotic mitochondria is linked to membrane pores . <i>EMBO J.</i> DOI 10.15252/embj.201593384 Winterflood, C.M. et al. (2015). Dual-Color 3D Superresolution Microscopy by Combined Spectral-Demixing and Biplane Imaging . <i>Biophys J.</i> 109, 3–6. Zhang, Z., et al. (2015). Ultrahigh-throughput single-molecule spectroscopy and spectrally resolved super-resolution microscopy . <i>Nature Methods</i> doi:10.1038/nmeth.3528
CF™680R	680/701	140,000	STED, single-molecule spectroscopy	Görlitz, F. et al. (2014). A STED Microscope Designed for Routine Biomedical Applications . <i>Progress Electromagnetics Res</i> 147, 57-68. König, I. et al. (2015). Single-molecule spectroscopy of protein conformational dynamics in live eukaryotic cells . <i>Nature Methods</i> doi:10.1038/nmeth.3475
CF™750	755/777	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; STED: Stimulated emission depletion; STORM: Stochastic optical reconstruction microscopy; TIRF: Total internal reflection fluorescence

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CF™ dyes and conjugates have been cited in hundreds of publications, with new references published every day. Visit www.biotium.com/downloads for a more comprehensive list of CF™ dye references by color and application.

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- Labeling is covalent, suitable for multiplex staining
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- Reactions tolerate common antibody buffer components and stabilizer proteins

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Label	Ex/Em (nm)	SE Protein Labeling Kits	VivoBrite Antibody Labeling Kits	Mix-n-Stain Antibody Labeling Kits			Mix-n-Stain Small Ligand Labeling Kits	
				1 labeling of 5-20 ug antibody	1 labeling of 20-50 ug antibody	1 labeling of 50-100 ug antibody	For cell surface targets	For intracellular targets
Biotin	N/A	92224		92286	92266	92244		
FITC	494/519			92294	92295	92296		
CF™350	347/448	92210		92270	92250	92230		
CF™405S	404/431	92211		92271	92251	92231		
CF™405M	408/452	92212		92272	92252	92232	92362	
CF™408	408/450							92356
CF™405L	395/495	92228		92303	92304	92305		
CF™488A	490/515	92213		92273	92253	92233	92350	
CF™500	500/510							92357
CF™514	516/548			92231	92232	92233		
CF™532	527/558	92208		92289	92290	92291		
CF™540	540/570							92358
CF™543	541/560	92209		92287	92267	92247		
CF™555	555/565	92214		92274	92254	92234		92364
CF™568	562/583	92215		92275	92255	92235	92351	
CF™594	593/614	92216		92276	92256	92236	92352	
CF™633	630/650	92217		92277	92257	92237	92353	
CF™640R	642/662	92225		92278	92258	92245	92354	
CF™647	650/665	92218		92279	92259	92238	92359	
CF™650	650/670							92363
CF™660C	667/685	92219		92280	92260	92239	92360	
CF™660R	663/682	92223		92281	92261	92243		
CF™680	681/698	92220	92160	92282	92262	92240	92361	
CF™680R	680/701	92226		92283	92263	92246	92355	
CF™750	755/777	92221	92161	92284	92264	92241		
CF™770	770/797	92222	92162	92285	92265	92242		
CF™790	784/806		92163	92288	92268	92248		
R-PE	496,564/578				92298**	92299		
APC	650/660				92306**	92307		
Per-CP	482/678				92308**	92309		
APC-CF750T	650/780				92310**	92311		
HRP	N/A			92300*	92301**	92302		
Alk. Phos.	N/A				92314**	92315		
Glucose oxidase	N/A				92312**	92313		
digoxigenin				92328	92329	92330		
DNP				92325	92326	92327		

*10-20 ug labeling; **25-50 ug labeling

CF™ Dye Reactive Dyes

A wide selection of colors and functional groups for dye conjugation

Reactive group/ unit size	Alkyne 0.5 mg	Amine 1 mg	Aminoxy 1 mg	Azide 0.5 mg	BCN 0.5 mg	Hydrazide 1 mg	Maleimide 1 umol	MTS 1 mg	Picolyl azide 0.5 mg	SE 1 umol	Tyramide** 0.5 mg
Reacts with	Azides, picolyl azides	Activated carboxylic acids	Aldehydes & ketones	Alkynes, BCN	Azides	Polar tracer*	Thiols	Thiols	Alkynes	Primary amines; lysine residues	HRP substrate
CF™350		92035	92050			92151	92020			92109	92170
CF™405S		92036	92055		92113	92183	92030			92110	92197
CF™405M	92093		92056	92092	92114		92021			92111	
CF™405L							92046			92112	92198
CF™430							92118			92117	
CF™440							92124			92123	
CF™450							96012			96011	
CF™488A	92086	92037	92051	92080	92075	92152	92022	92097	92187	92120	92171
CF™500					96026						
CF™514										92103	92199
CF™532				92180			92045			92104	
CF™543				92181			92044	92098		92105	92172
CF™555	92087	92038		92081		92153	92023			92130	96021
CF™568	92088	92039	92057	92082	92076	92154	92024		92188	92131	92173
CF™570							96015			96014	
CF™583							96017			96016	
CF™594	92089	92040	92052	92083	92077	92158	92025	92099	92189	92132	92174
CF™620R							92033			92106	92194
CF™633		92041	92053			92156	92026			92133	
CF™640R	92091	92043	92058	92085	92078	92157	92034	92096	92190	92108	92175
CF™647	92090	92042		92084		92136	92027		92191	92135	96022
CF™650					96027						
CF™660C	92095			92094			92028		96001	92137	
CF™660R	96004	96010	92059	92182		96024	92031			92134	92195
CF™680	96005			92119			92029		96003	92139	
CF™680R	96006		92054		92079	96025	92032		96007	92107	92196
CF™750		92102								92142	
CF™770		92065				92192				92150	
CF™790										92155*	
CF™800							92128			92127*	

* For conjugation to aldehyde or ketone groups, we recommend using CF™ dye aminoxy forms.

* Unit size 0.25 umol

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CF™ Dye Bioconjugates

Bioconjugate applications

Conjugate	Application
Annexin V	Phosphatidyl serine probe; apoptosis marker
α-Bungarotoxin	Acetylcholine receptor probe; neuromuscular junction stain
Bovine serum albumin (BSA)	Fluid-phase endocytosis tracer; in vivo blood flow tracer
Cholera Toxin Subunit B	GM1 receptor probe; lipid raft, endocytic vesicle, neuronal tracing
Concanavalin A (Con A)	Lectin; binds α-D-mannosyl and α-D-glucosyl groups, stains yeast cell wall
Dextran, anionic, fixable: MW 10K, 40K, 150K, 250K	Fluid-phase endocytosis tracer
Phalloidin	Filamentous actin probe
Peanut agglutinin (PNA)	Lectin; specific for terminal β-galactose
Streptavidin	Detection of biotinylated probes
Transferrin (human)	Recycling endosome tracer
Wheat germ agglutinin (WGA)	Lectin, binds N-acetyl-D-glucosamine and sialic acid; bacterial Gram stain, stains yeast bud scars

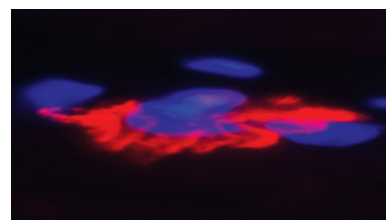


Figure 1. Frozen section of rat skeletal muscle stained with CF633 α-bungarotoxin (magenta) to detect nicotinic acetylcholine receptors at the neuromuscular junction. Nuclei are stained with DAPI (blue).

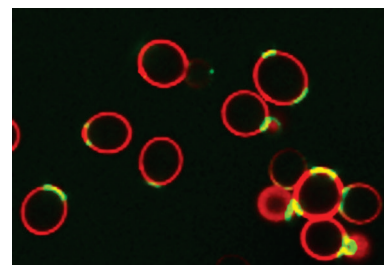


Figure 2. *S. cerevisiae* yeast stained with CF488A WGA and CF594 ConA. ConA (red) stains the cell wall, while WGA (green) preferentially stains bud scars.

CF™ dye bioconjugates

Conjugate	Annexin V	α-Bungarotoxin	BSA	Cholera Toxin B	Con A	Dextran 10,000 MW	Phalloidin	PNA	Streptavidin	Transferrin	WGA
CF™350	29012				29015		00049		29031		29021
CF™405S		00002			29075				29032		29027
CF™405M	29009				29074		00034		29033		29028
CF™405L									29056		
CF™430							00054		29065		
CF™440							00055		29066		
CF™488A	29005	00005	20289	00070	29016	80110	00042	29060	29034	00081	29022
CF™532				00074			00051		29030		29064
CF™543		00026		00075		80111	00043		29043	00082	
CF™555	29004	00018				80112	00040		29038		29076
CF™568	29010	00006		00071		80113	00044	29061	29035	00083	29077
CF™594	29011	00007	20290	00072	29017	80114	00045	29062	29036	00084	29023
CF™620R				00076							
CF™633	29008	00009		00077	29018		00046		29037		29024
CF™640R	29014	00004	20291	00073	29019	80115	00050	29063	29041	00085	29026
CF™647	29003						00041		29039		
CF™660C							00052				
CF™660R	29069			00078			00047		29040		
CF™680	29007		20292		29020	80118	00053				29029
CF™680R	29070	00003		00079		80116	00048		29042	00086	29025
CF™750	29006					80119				00087	
CF™770	29046				29058	80120					29059
CF™790	29047					80121					

Visit www.biotium.com to see our selection of apoptosis staining kits, bacterial Gram stain kits, and phalloidin conjugates of biotin and traditional dyes.

Nucleotide conjugates for probe synthesis and TUNEL assay

	CF™405S	CF™488A	CF™532	CF™543	CF™555	CF™568	CF™594	CF™640R	CF™647	CF™660R	CF™680R
dCTP		40067	40057	40058	40027	40055	40056	40066	40028	40068	
ddCTP					40031						
UTP								40032			
dUTP	40004	40008		40002		40005	40006	40007			40003

Visit www.biotium.com to see our CF™ dye TUNEL staining kits, plus a selection of nucleotide conjugates of biotin, traditional dyes, and amino-allyl nucleotides.

Primary antibody conjugates

Our collection of monoclonal primary antibodies is constantly growing, visit www.biotium.com to see the most up-to-date offerings. Available in the following formats:

- Purified: 200 ug/mL in PBS with 0.05% BSA & 0.05% azide, 100 uL (20 ug) or 500 uL (100 ug) unit size
- Purified, BSA-free: 1 mg/ml in PBS with 0.05% azide, ready-to-use for Mix-n-Stain™ labeling (see p. 23) or other conjugation, 50 uL (50 ug) unit size
- Conjugates: Your choice of 12 CF™ dye colors for microscopy, flow cytometry, or near-infrared western blot, plus R-PE, APC, and PerCP; 100 ug/mL in PBS with 0.05% BSA & 0.05% azide, 100 uL (10 ug) or 500 uL (50 ug) unit size

A	CD27	Complement C4d	HCG-beta	MHC II	Prostate Specific Antigen
A, Forssman	CD28	Creatine Phosphokinase	HCG-intact	Milk Fat Globulin	Proximal Nephrogenic
ACTH	CD282 / TLR2	Cyclin A2	Helicobacter pylori	MiTF	Antigen
Adenosine Monophosphate	CD284 / TLR4	Cyclin B1	Heparan Sulphate Proteoglycan	Mitochondria	pS2
Deaminase 3	CD30	Cyclin D1	Hepatocyte Specific Antigen	Mitochondrial Marker	PSA
AFP	CD31 / PECAM-1	Cytochrome C	HepPar-1	Moesin	PTEN
Alkaline Phosphatase	CD32	Cytokeratin 10	HER-2 / CD340	MRP-14	PTH
AMACR / p504S	CD33	Cytokeratin 10/13	HIF1?	MUC18 / Mucin 18 / CD146	PTH / Parathyroid
Androgen Receptor	CD34	Cytokeratin 14	Histiocytoma Marker	MUC2	Hormone
Arginase 1	CD35 / CR1	Cytokeratin 17	Histone H1	MUC5AC	
	CD36	Cytokeratin 18	HLA-Aw32 & HLA-A25	Mucin 1 / EMA / Episialin /	R
B	CD37	Cytokeratin 19	HLA-B	CD227	Rabies
Bax	CD38	Cytokeratin 5/8	HLA-DRB	Mucin 3	Retinol Binding Protein-1
BCL-10	CD3e	Cytokeratin 6	HSP27	Mucin 5AC	
bcl-2	CD4	Cytokeratin 7	HSP60	Mucin 6	S
bcl-6	CD41a	Cytokeratin 8	Human Nuclear Antigen	Muscle Specific Actin	S100
bcl-x	CD43	Cytokeratin 8/18	Human Nucleolar Antigen	Myeloid-Associated	S100A9
Beta Catenin	CD44 Standard	Cytokeratin, Acidic	Human Papillomavirus 16	Differentiation Marker	SHBG
Beta-2 Microglobulin	CD45 / LCA	Cytokeratin, Basic		MyoD1	Small Cell Lung Cancer
Biotin	CD45RA	Cytokeratin, HMW		Myogenin	Smooth Muscle Actin
Blood Group A	CD45RB	Cytokeratin, LMW		Myosin, Smooth Muscle Heavy	SOX10
Blood Group Antigen A	CD45RO	Cytokeratin, multi		Chain	SUMO-1
Blood Group Antigen B	CD46	Cytokeratin, pan			SUMO-2
Bovine Serum Albumin	CD47		I		SUMO-2/3
BrdU	CD48		IDH1		
Bromodeoxyuridine	CD5	D	IgA Immunoglobulin		
	CD50	DOG-1	IgA Secretory Component		
	CD53	Double Stranded DNA	IGF-1		
	CD54		IgG	N	
C	CD54 / ICAM-1	E	IgG Immunoglobulin	Napsin-A	
CA19-9	CD55	E-Cadherin / CD324	IgM Immunoglobulin	Neurofilament	
Caldesmon, HMW	CD56 / NCAM	EGFR	IL-6	Neurofilament, phospho	T
Calgranulin B	CD57 / B3GAT1	EMI1	Insulin	NGFR	TAG-72 / CA72.4
Calponin-1	CD59	Eosinophil Peroxidase	Interferon alpha 1	NKX2.2	Tenascin
Calprotectin	CD6	Ep-CAM / CD326	Interferon alpha-2	Nuclear Antigen	Testosterone
Carbonic Anhydrase IX	CD61	Erythrocyte Specific	Interferon gamma	Nuclear Membrane	TGFalpha
Carcinoembryonic Antigen	CD63	Estrogen Receptor	Involucrin	Nuclear Membrane Marker	TGF-beta
CD10	CD66	Estrogen Receptor beta 1	IPO-38	Nucleolar / Nucleoli	Thomsen-Friedenreich Ag.
CD100	CD66, pan		Isotype control, mouse IgG1, κ	Nucleolin	Thymidylate Synthase
CD104	CD68		Isotype control, mouse IgG2a, κ	NuMA	Thyroglobulin
CD106 / VCAM1	CD7	F	Isotype control, mouse IgG2b, κ		TIMP3
CD117	CD70	Fascin-1		O	TNF alpha
CD11a	CD71	FGF23		ODC-1	Topoisomerase I, MT
CD11b	CD74	Fibronectin	K		TOX3
CD11c	CD79a	FOXP3	Kappa Light Chain		TRAcP
CD13	CD8	FSH beta	Ksp-Cadherin / CDH16	P	Transgelin / SM22-alpha
CD14	CD84		Ku-Holo	p21 / WAF1	Transglutaminase II
CD146 / Mucin 18	CD86	G		p24-HIV	TRIM29
CD147	CD8A	GFAP		p27 / KIP1	TRP1
CD15 / FUT4 / Lewis x	CD8B	GITR / Tnfrsf18		p34 / cdk1	TTF-1 / NKX2.1
CD16	CD89	GLG1	L	p40	Tyrosinase
CD16 / Fc-gamma Receptor III	CD90 / Thy1	Glucose Regulated Protein 94	Lambda Light Chain	p53	
CD171 / L1CAM	CD95	Glycophorin A / CD235a	Laminin	p55;50 EBV-Early Antigen	U
CD176 / T-F Ag	CD98	Glypican-3	LEC Chemokine	p57 / KIP2	UACA / Nucling
CD18	CD99	GM-CSF	Lewis A	p57Kip2	UGT1A9
CD19	CD99	GnRH-Receptor	Lewis B	PAX6	UPK3A
CD195 / CCR-5	Cdc20	Golgi Complex	Liver Canaliculi	PAX7	
CD1a	CDw17	gp100 / Melanosome	Lung Specific Antigen	PCNA	V
CD1b	CDw60	Granulocyte Marker	Luteinizing Hormone beta	PD1 / PDCD1 / CD279	VEGF-A
CD2	CDw75	GCSF		pgp9.5	VEGFR1 / Fit-1
CD20	CELA3B	Granzyme B	M	Phosphotyrosine	VEGFR2 / Fik-1
CD21	Chromogranin A		Macrophage Specific Antigen	PLAP	Vimentin
CD22	CMV-p65		MAGE A1	Plasma Cell Marker	
CD25	c-Myb		Major Vault Protein	PLGF	
CD26	c-Myc		MALT-1	Pmel17 / gp100 / SILV	W
			MAP3K1	Podocalyxin	von Willebrand Factor
			MART-1 / Melan-A	Progesterone	WT1
			MCAM / MUC18 / Mucin 18 / CD146	Progesterone Receptor	
			Melanoma Marker	Prolactin Receptor	Z
			MHC I		ZAP70

CF™ Dye Anti-Tag and Secondary Antibody Conjugates

Anti-GFP, anti-hapten, and anti-epitope tag antibody conjugates

In PBS, 50% glycerol, 2 mg/ml BSA, 0.05% sodium azide

	Goat anti-GST	Mouse monoclonal anti-biotin	Mouse monoclonal anti-fluorescein	Mouse monoclonal anti-GFP	Mouse monoclonal anti-6X His tag	Rabbit anti-HA tag	Rabbit anti-RFP
Concentration/ Unit size	1 mg/mL 0.1 mL	2 mg/mL 50 uL or 0.25 mL	2 mg/mL 50 uL or 0.25 mL	1 mg/mL 0.1 mL	1 mg/mL 0.1 mL	1 mg/mL 50 uL	1 mg/mL 0.1 mL
CF™405S		20203					
CF™405M			20214				
CF™488A	20424	20204	20210	20215	20228	20238	20421
CF™594	20425	20205	20211	20216	20229	20239	20422
CF™633		20206	20212	20217			
CF™640R	20426	20207	20213	20218	20237	20237	20423
CF™647							
CF™680				20219	20359		
CF™770		20367		20220	20360		

Secondary antibodies, whole IgG (H+L), not highly cross-adsorbed

2 mg/mL in PBS, 50% glycerol, 2 mg/ml BSA, 0.05% sodium azide, or preservative-free lyophilized form
Unit size: 0.5 mL, 50 uL, or 1 mg (lyophilized)

	Chicken anti-goat	Chicken anti-mouse	Chicken anti-rabbit	Goat anti-Guinea pig	Goat anti-mouse	Goat anti-rabbit	Goat anti-swine	Llama anti-mouse	Llama anti-rabbit	Rabbit anti-chicken	Rabbit anti-goat	Rabbit anti-guinea pig
CF™350	20364	20331	20332	20198	20140	20141						
CF™405S					20080	20082						
CF™405M					20180	20181						
CF™405L					20408	20409						
CF™488A	20225	20208	20209	20017	20010	20012	20028	20454	20449	20079	20021	
CF™514					20386	20387						
CF™532					20365	20366						
CF™543	20333	20334	20335	20317	20306	20309	20324			20312	20315	20336
CF™555				20036	20030	20033	20236				20031	
CF™568	20337	20338	20339	20108	20100	20102	20091	20455	20450		20107	
CF™594	20226	20221	20223	20118	20110	20112	20160	20456	20451	20164	20117	
CF™633	20227	20222	20224	20129	20120	20122	20138			20165	20128	
CF™640R				20085	20197	20202	20089	20457	20452			
CF™647				20041	20040	20043	20286	20458	20453			
CF™660C					20050	20053						
CF™660R					20054	20055						
CF™680											20068	20243
CF™750					20070	20073						
CF™770												20244
CF™790					20378							

Don't see what you're looking for?

We regularly add new CF™ dye conjugates to our catalog according to customer demand. Be sure to check our website for updates. If you are looking for a CF™ dye product not listed in our catalog, please let us know. We may be able to add it as a new product, or perform a custom conjugation for you.

Visit www.biotium.com to see our full selection of secondary antibodies, including conjugates of biotin, HRP, R-PE, and APC.

CF™ Dye Secondary Antibody Conjugates

Highly cross-adsorbed for multiple labeling

Drop-n-Stain™ secondary antibodies, whole IgG (H+L), highly cross-adsorbed

5 mL solution in convenient dropper bottle format for quick and easy immunofluorescence staining.

	Donkey anti-mouse	Donkey anti-rabbit	Goat anti-mouse	Goat anti-rabbit
Min x react	Bv, Ch, Gt, GP, Hs, Hu, Rb, Sh, SHm	Bv, Ch, Gt, GP, Hs, Hu, Ms, Rt, Sh, SHm	Bv, Hs, Hu, Rb, Sw	Hu, Ms, Rt
CF™488A	20952	20950	20956	20954
CF™543	20967	20966	20969	20968
CF™594	20953	20951	20957	20955
CF™640R	20963	20962	20965	20964

Secondary antibodies, whole IgG (H+L), highly cross-adsorbed

2 mg/mL in PBS. 50% glycerol, 2 mg/ml BSA, 0.05% sodium azide, or preservative-free lyophilized form
CF™350-CF™660R unit sizes: 0.5 mL, 50 uL, or 1 mg (lyophilized); CF™680-CF™790 available in 0.25 mL or 50 uL sizes

	Bovine anti-goat	Donkey anti-chicken	Donkey anti-goat	Donkey anti-guinea pig	Donkey anti-human	Donkey anti-mouse	Donkey anti-rabbit	Donkey anti-rat	Donkey anti-sheep
Min x react	Bv, Ch, GP, Hs, Hu, Ms, Rb, Rt, SHm	Bv, Gt, GP, Hs, Hu, Ms, Rb, Rt, Sh, SHm	Ch, GP, Hs, Hu, Ms, Rb, Rt, SHm	Bv, Ch, Gt, Hs, Hu, Ms, Rb, Sh, SHm	Bv, Ch, GP, Gt, Hs, Ms, Rb, Rt, Sh, SHm	Bv, Ch, Gt, GP, Hs, Hu, Rb, Sh, SHm	Bv, Ch, Gt, GP, Hs, Hu, Ms, Sh, SHm	Bv, Ch, GP, Gt, Hs, Hu, Ms, Rb, Sh, SHm	Ch, GP, Hs, Hu, Ms, Rb, Rt, SHm
CF™350		20275	20142			20350	20351	20361	20148
CF™405S				20356			20420	20419	
CF™405M				20376					
CF™430						20461	20462		
CF™488A	20293	20166	20016	20169	20074	20014	20015	20027	20024
CF™543	20313	20310	20314	20316	20318	20305	20308	20320	20322
CF™555			20039	20276		20037	20038		20234
CF™568	20294		20106	20377		20105	20098	20092	20095
CF™594	20295	20167	20116	20170	20075	20115	20152	20159	20156
CF™633	20296	20168	20127	20171	20076	20124	20125	20137	20134
CF™640R	20297		20179			20177	20178	20199	20083
CF™647			20048			20046	20047		20284
CF™660C			20051	20372					
CF™660R						20388	20389	20390	
CF™680			20060	20241	20278		20418	20417	20062
CF™680R			20196			20194	20195		
CF™750			20362				20298		
CF™770			20277	20242					
CF™790			20345		20279	20363	20344		

Bv: bovine; Ch: chicken; Gt: goat; GP: guinea pig; Hs: horse; Hu: human; Ms: mouse; Rb: rabbit; Sh: sheep; SHm: Syrian hamster; Sw: swine; Rt: rat

See more highly cross-adsorbed secondaries on the next page

Don't see what you're looking for?

We regularly add new CF™ dye conjugates to our catalog according to customer demand. Be sure to check our website for updates. If you are looking for a CF™ dye product not listed in our catalog, please let us know. We may be able to add it as a new product, or perform a custom conjugation for you.

Visit www.biotium.com to see our full selection of secondary antibodies, including conjugates of biotin, HRP, R-PE, and APC.

CF™ Dye Secondary Antibody Conjugates

Highly cross-adsorbed, F(ab')₂ fragments, and isotype-specific secondary antibodies

Secondary antibodies, whole IgG (H+L) , highly cross-adsorbed (continued from p. 28)

2 mg/mL in PBS. 50% glycerol, 2 mg/ml BSA, 0.05% sodium azide, or preservative-free lyophilized form
CF™350-CF™660R unit sizes: 0.5 mL, 50 uL, or 1 mg (lyophilized); CF™680-CF™790 available in 0.25 mL or 50 uL sizes

	Goat anti-chicken	Goat anti-human	Goat anti-mouse	Goat anti-mouse (min x rat)	Goat anti-rabbit	Goat anti-rat	Rabbit anti-human	Rabbit anti-mouse	Rabbit anti-rat	Rabbit anti-sheep
	Bv, Gt, GP, Hs, Hu, Ms, Rb, Rt, Sh, SHm	Bv, Hs, Ms	Bv, Hs, Hu, Rb, Sw	Bv, Ch, Gt, GP, Hs, Hu, Rb, Rt, Sh, SHm	Hu, Ms, Rt	Bv, Hs, Hu, Rb	Ms	Hu	Hu	Hu
CF™350			20143		20144	20147		20149		
CF™405S										
CF™405M	20375		20182	20340	20373	20374				
CF™430			20459		20460					
CF™488A	20020	20022	20018	20302	20019	20023	20071	20026	20025	20172
CF™543	20311	20319	20299	20328	20300	20321		20307		20323
CF™555	20034	20320	20231		20232	20233		20235		
CF™568	20104	20097	20101	20301	20103	20096		20093	20094	
CF™594	20114	20154	20111	20303	20113	20155	20072	20158	20157	20173
CF™633	20126	20132	20121	20341	20123	20133	20066	20136	20135	20174
CF™640R	20084	20081	20175	20304	20176	20088		20200	20201	
CF™647	20044	20280	20281		20282	20283		20285		
CF™660C	20371		20052	20356	20369	20370				
CF™660R										
CF™680		20287	20065		20067	20069		20061		
CF™680R			20192		20193					
CF™750			20463							
CF™770		20288	20077		20078	20383				
CF™790			20342		20343					

Bv: bovine; Ch: chicken; Gt: goat; GP: guinea pig; Hs: horse; Hu: human; Ms: mouse; Rb: rabbit; Sh: sheep; SHm: Syrian hamster; Sw: swine; Rt: rat

Secondary antibodies, F(ab')₂ fragments

2 mg/mL, unit size: 0.25 mL or 50 uL

	Goat anti-mouse, F(ab') ₂	Goat anti-rabbit, F(ab') ₂
CF™350	20145	20146
CF™488A	20011	20013
CF™543	20329	20330
CF™555	20032	20035
CF™568	20109	20099
CF™594	20119	20153
CF™633	20130	20131
CF™640R	20086	20087
CF™647	20042	20045
CF™680	20063	20064

Goat anti-mouse isotype-specific antibodies

Highly cross-adsorbed for multiple labeling (min X Bv, Hu, Rb)
2 mg/mL, unit size: 0.25 mL or 50 uL

	Goat anti-mouse IgG1	Goat anti-mouse IgG2a	Goat anti-mouse IgG2b
CF™350	20245	20255	20265
CF™405S	20380	20381	20382
CF™488A	20246	20256	20266
CF™543	20325	20356	20327
CF™555	20247	20257	20267
CF™568	20248	20258	20268
CF™594	20249	20259	20269
CF™633	20250	20260	20270
CF™640R	20251	20261	20271
CF™647	20252	20262	20272
CF™680	20253	20263	20273
CF™770	20254	20264	20274

Goat anti-human isotype-specific antibodies

2 mg/mL, unit size: 0.25 mL or 50 uL

	Goat anti-human IgA (alpha chain)	Goat anti-human IgM (mu chain)
CF™488A	20428	20347
CF™594	20429	20348
CF™640R		20349
CF™633	20427	
CF™647		20346
CF™680		20384
CF™770		20385

Related Products and Accessories

Buffers, mounting media, counterstains and more

RedDot™ 1 and RedDot™ 2 Far-Red Nuclear Stains

RedDot™ 1

- Replaces Draq®5 for live cell nuclear staining
- Cell normalization for In Cell Western®
- Cell cycle analysis by flow cytometry

RedDot™ 2

- Replaces Draq®7 for dead cell staining and fixed cell staining
- More specific than Draq®7 for fixed cell nuclear counterstaining

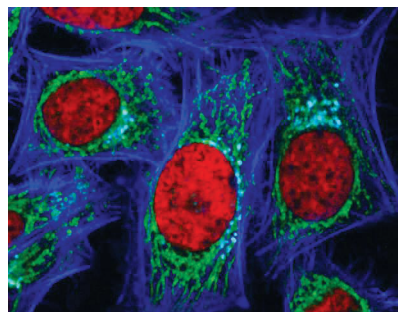
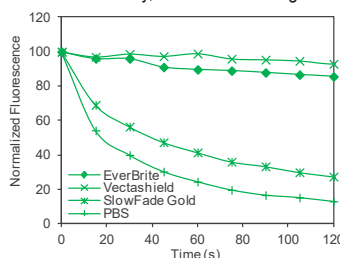


Figure 1. HeLa cells stained with rabbit anti-COX IV and CF488A goat anti-rabbit (mitochondria, green), and mouse anti-Golgin 97 and CF555 goat anti-mouse (Golgi, cyan) Actin filaments are stained with CF405 phalloidin (blue) and nuclei are stained with RedDot2 (red).

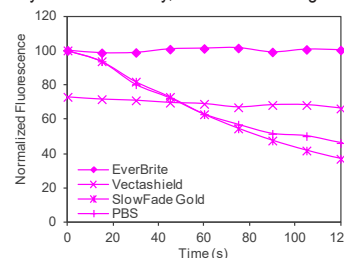
EverBrite™ Mounting Media

- Superior antifade protection
- Compatible with CF™ dyes and other dyes
- Compatible with cyanine-based dyes (Cy® dyes, Alexa Fluor® 647 and DyLight® 649), unlike VectaShield®
- Available in wet-set or hardset formulations
- With or without DAPI

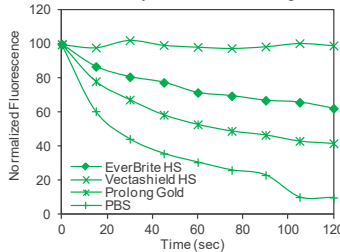
FITC Photostability, Wet-Set Mounting Media



Cy5 Photostability, Wet-Set Mounting Media



FITC Photostability, Hardset Mounting Media



Cy5 Photostability, Hardset Mounting Media

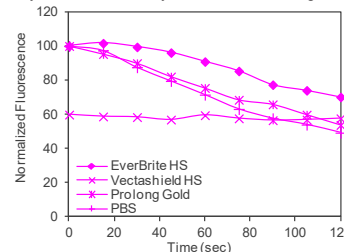


Figure 2. Photostability of HeLa cell immunofluorescence staining in various mounting media. A major advantage of EverBrite™ mounting medium is that, unlike VectaShield®, it does not decrease the fluorescence of cyanine-based fluorophores. Fluorescence values for Cy®5 in VectaShield® media are normalized to PBS time 0 to illustrate the drop in fluorescence of cyanine dyes caused by VectaShield®.

CoverGrip™ Coverslip Sealant

- Designed specifically for sealing coverslip edges
- Won't mix with aqueous mounting medium like nail polish can
- Made with natural Limonene solvent
- 15 mL brush bottle, or 100 mL refill



Figure 3. CoverGrip Coverslip Sealant brush bottle and refill bottle.

Super^{HT} Pap Pens

- Create hydrophobic barriers on glass slides to separate specimens and conserve antibodies
- Insoluble in aqueous buffers, detergents, alcohol, or acetone, removable with xylene
- Stable at temperatures up to 120°C
- Super^{HT} Mini Pap Pen: 2.5 mm tip, ~400 applications
- Super^{HT} Pap Pen: 4 mm tip, ~800 applications



Figure 4. Super^{HT} Pap Pens.

Related Products and Accessories

TrueBlack™ Lipofuscin Autofluorescence Quencher

- Eliminates autofluorescence from lipofuscin in human and aged animal tissue sections
- Reduces autofluorescence from other sources, such as red blood cells and collagen/elastin
- Less red/far-red background compared to Sudan Black B
- Can be used to treat tissue sections before or after antibody staining

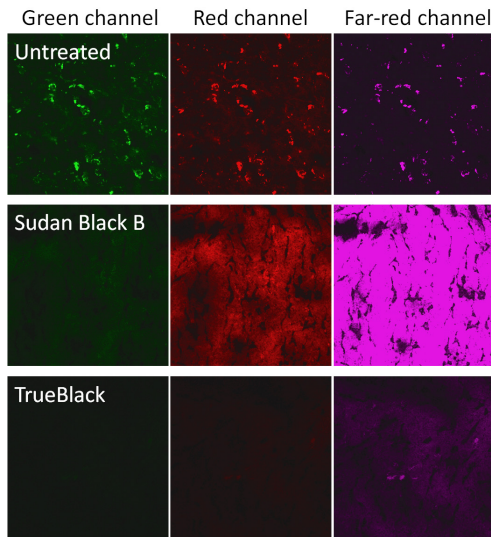


Figure 3. Lipofuscin autofluorescence in human cerebral cortex sections. Top row: lipofuscin appears as highly fluorescent granules throughout the tissue section in all three fluorescent channels. Middle row: Sudan Black B masks lipofuscin autofluorescence, but causes high background in the red and far-red channels. Bottom row: TrueBlack quenches lipofuscin with minimal increase in fluorescence background (bottom row).

Accessory Products

Catalog No.	Product Name
23001	EverBrite™ Mounting Medium
23002	EverBrite™ Mounting Medium with DAPI
23003	EverBrite™ Hardset Mounting Medium
23004	EverBrite™ Hardset Mounting Medium with DAPI
23005	CoverGrip™ Coverslip Sealant
23007	TrueBlack™ Lipofuscin Autofluorescence Quencher, 20X in DMF
40060	RedDot™ 1 Far Red Nuclear Counterstain, 200X in H ₂ O
40061	RedDot™ 2 Far Red Nuclear Counterstain, 200X in DMSO
30069	AccuEasy™ Flow Cytometry Kit
22003	Mini Cell Scrapers
22005	Mini Super ^{HT} Pap Pen
22006	Super ^{HT} Pap Pen
23006	Flow Cytometry Fixation/Permeabilization Kit
22015	Fixation Buffer
22016	Permeabilization Buffer
22017	Permeabilization and Blocking Buffer (5X)
22010	10X Fish Gelatin Blocking Agent

AccuEasy™ Flow Cytometry Kit

- Stain and harvest adherent cells for flow cytometry
- Prevents loss of surface marker staining upon cell detachment
- Increases sensitivity of cell surface marker detection compared to conventional methods

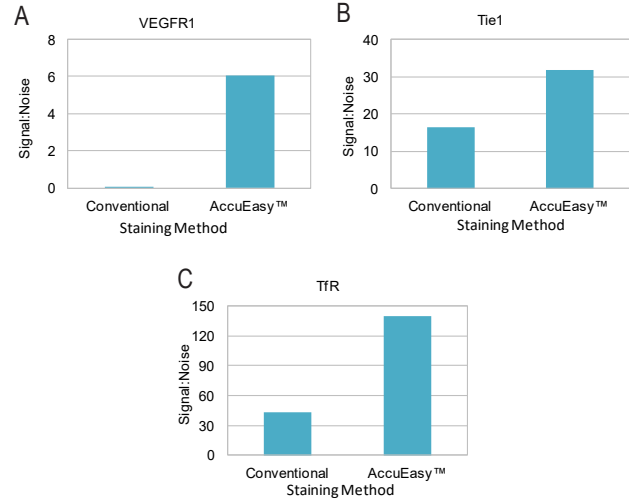


Figure 1. The AccuEasy staining method improves signal to noise ratio in flow cytometry analysis of cell surface markers on adherent cells compared conventional methods. A. Indirect immunofluorescence staining of EA.hy926 human endothelial cells with mouse monoclonal antibody against VEGF Receptor 1 (VEGFR1) or isotype control and PE conjugated anti-mouse IgG. B. Staining of EA.hy926 cells with mouse monoclonal antibody against Tie1 or isotype antibody control and PE conjugated anti-mouse IgG. C. Direct immunofluorescence staining of HeLa cells with Mix-n-Stain™ CF™ 488A labeled mouse monoclonal antibody against transferrin receptor (TfR), or isotype control.

Mini Cell Scrapers

- For harvesting cells from 96-, 48-, or 24-well plates
- 0.5 cm wide and 6 cm long polystyrene scrapers
- Sterile and disposable

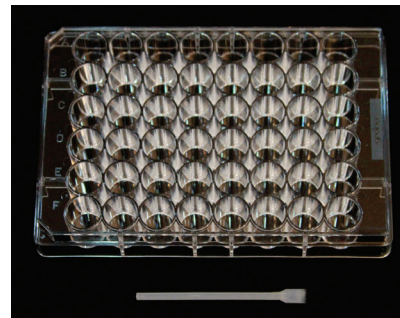
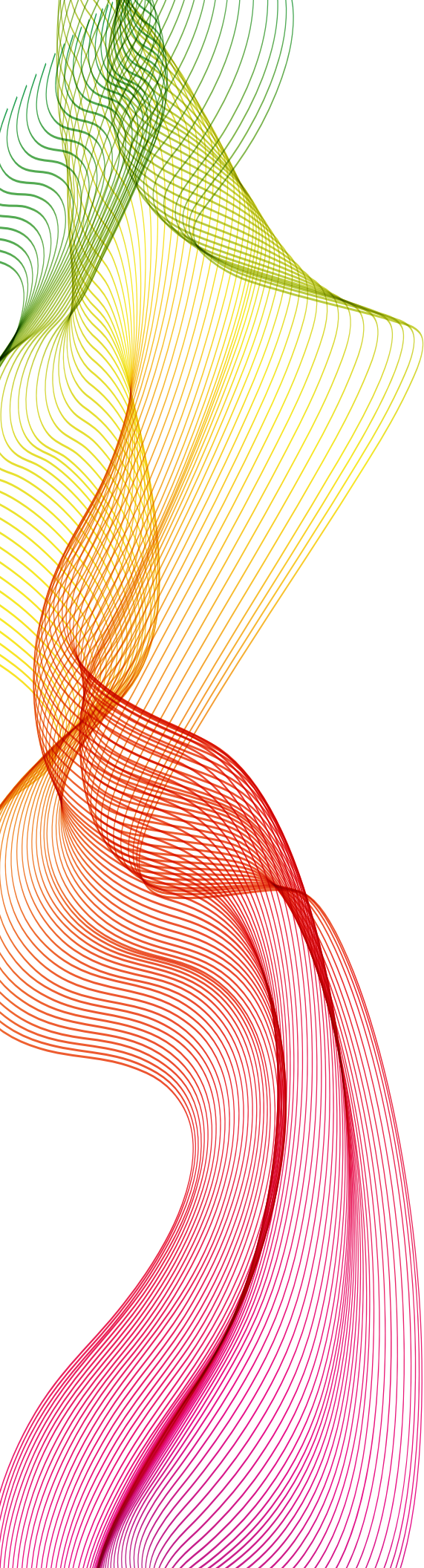


Figure 2. Mini Cell Scraper shown above next to a 48-well plate for scale.

Visit www.biotium.com to find more buffers, counterstains, and accessories.



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