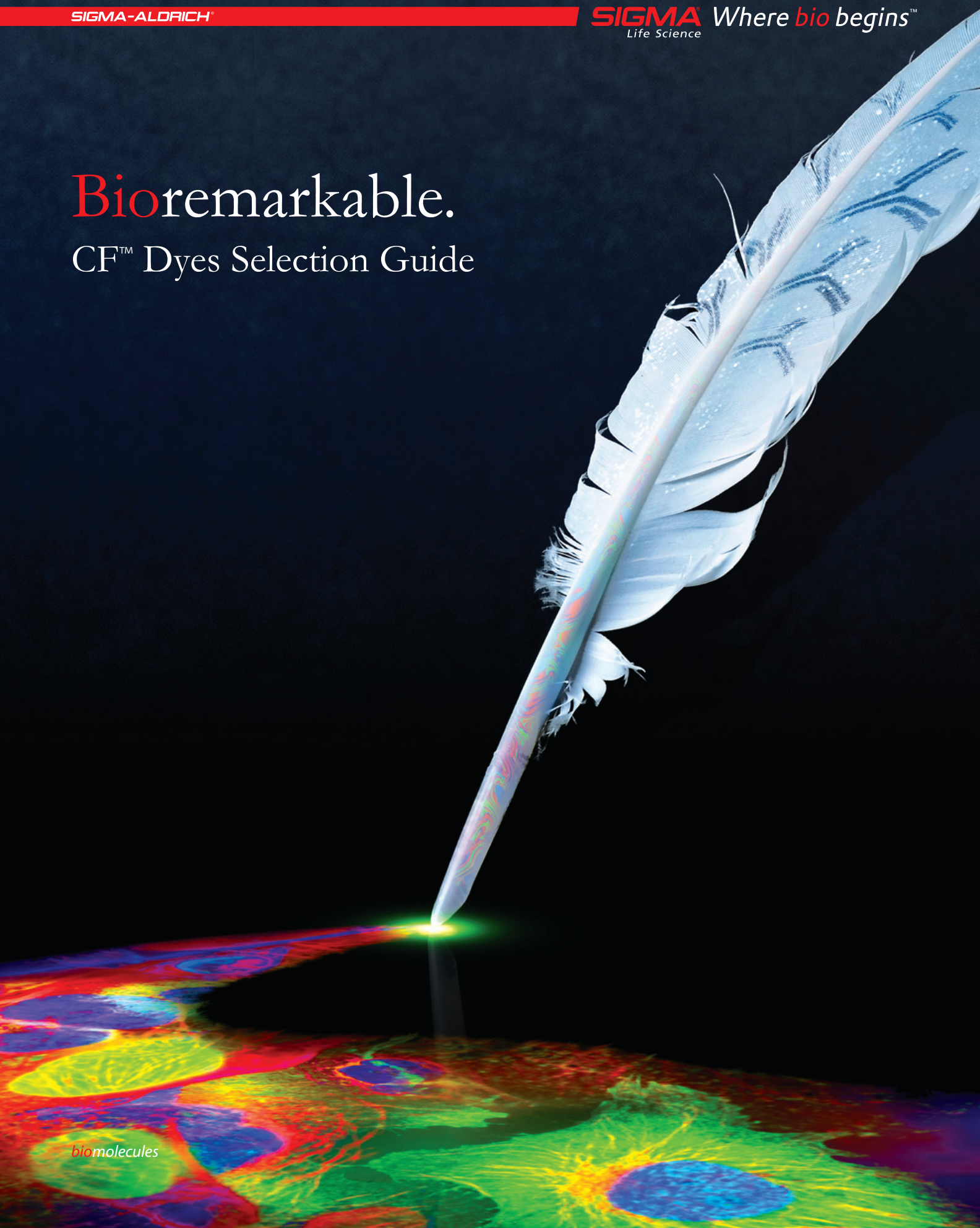
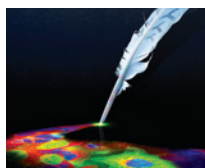


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CF™ Dyes Selection Guide





Bioremarkable
CF™ Dyes Selection Guide

CF Dyes

Achieve greater sensitivity, brighter results, and better photostability with enhanced CF dye technology.

CF dyes are a series of highly water-soluble fluorescent dyes spanning the visible and near-infrared (near-IR) spectrum (Figure 1A and 1B) for labeling antibodies, proteins, nucleic acids, and other biomolecules. Developed by scientists using new breakthrough chemistries, the brightness, photostability, and color selection of CF dyes rival or exceed the quality of other commercial dyes as a result of rational dye design.

Sigma® Life Science currently offers 19 CF dyes spanning the visible and near-IR wavelengths, with additional colors in development.

The CF dye product line includes reactive CF dyes, labeling kits, CF-labeled secondary antibodies, and other bioconjugates. This collection further expands Sigma Life Science’s broad range of carefully selected secondary antibodies and conjugates, allowing scientists to achieve greater sensitivity, brighter results, and better photostability in immunoassays.

The CF dye quick reference guide will help you choose the correct dye for your application.

Absorption/Emission Spectra of Goat Anti-Mouse Antibody

Figure 1A

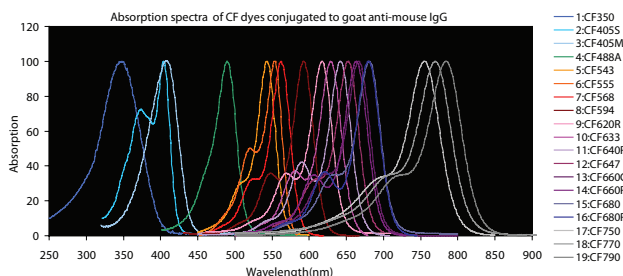
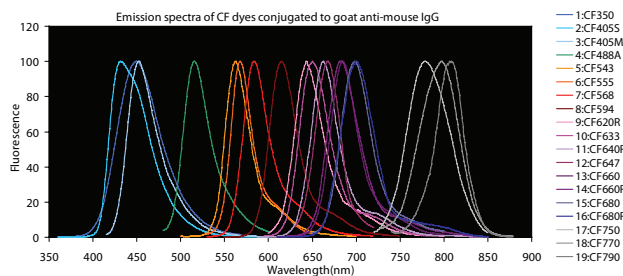
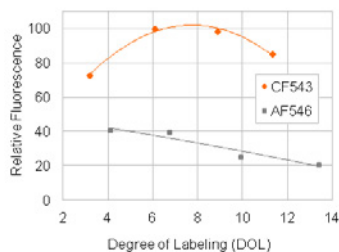


Figure 1B



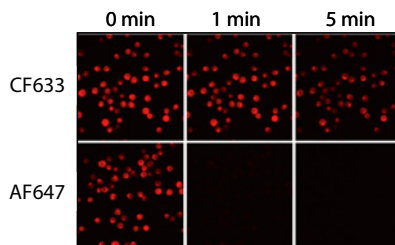
Brightness, Photostability and Conjugate Specificity of CF Dyes Compared to Other Commercial Dyes

Figure 2A



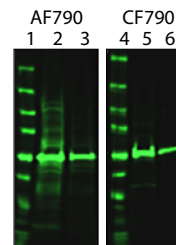
Relative fluorescence of CF543 and Alexa Fluor 546 (AF546) goat anti-mouse conjugates as a function of the number of dye molecules per protein (degree of labeling).

Figure 2B



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.

Figure 2C



Near-IR CF dyes are highly water soluble without carrying excessive negative charge, which can increase non-specific binding of antibody conjugates. Near-IR western blots imaged using the Odyssey system (Li-COR Biosciences). Two dilutions of HeLa cell lysate were probed with mouse anti-tubulin antibody followed by goat anti-mouse conjugated to Alexa Fluor 790 (AF790) (2, 3) or CF790 (5, 6).

Fluorescent Dye Comparison

CF™ dye	Reactive forms	λEx (nm)	λEm (nm)	MW	ε	Excitation source	Direct replacement for:	Reactive forms	λEx (nm)	λEm (nm)	MW	ε	Advantages of CF™ dye
CF™ 350	amine, aminoxy, hydrazide, maleimide, SE	347	448	496	18,000	UV	AMCA	NHS, sulfo-NHS, hydrazide, HPDP, SE	353	442	330	19,000	- Yields the brightest blue fluorescent antibody conjugates when excited at ~350 nm - Highly water-soluble and insensitive to pH
							Cascade Blue®	Acetyl azide, hydrazid, ethylenediamine	400	420	596	28,000	
							DyLight™ 350	NHS, maleimide	353	432	899	15,000	
CF™ 405S	amine, aminoxy, maleimide, SE	404	431	1169	33,000	405 nm laser	Alexa Fluor® 405	SE, cadaverine	400	424	1028	35,000	- Much brighter than Alexa Fluor® 405 due to better compatibility with excitation and emission windows on common instruments
							DyLight™ 405	NHS, maleimide	400	420	793	30,000	- Highly water-soluble and insensitive to pH
CF™ 405M	aminoxy, maleimide, SE	408	452	503	41,000	405 nm laser	BD Horizon™ V450	Conjugated	404	448			- More photostable than Pacific Blue® dye - As bright as Pacific Blue® dye in the blue channel
							eFluor® 450	Conjugated	405	450			- Less spill-over fluorescence in the green channel
							Pacific Blue®	SE	405	455	406	46,000	- Highly water-soluble
CF™ 488A	amine, aminoxy, hydrazide, maleimide, SE	490	515	914	70,000	488 nm laser	Alexa Fluor® 488	5-SDPE, 5-TFPE, maleimide, alkyne, azide, cadaverine, hydrazide, hydroxylamine, SE	494	517	643	73,000	- Minimal charge compared to Alexa Fluor® 488 reduces nonspecific antibody staining
							Cy 2	Bis NHS	489	506	714	150,000	- Less spill-over fluorescence in the red channel than Alexa Fluor® 488
							DY-495	NHS, maleimide, carboxylic acid, amino derivative	493	521	623	70,000	- Extremely photostable
							DyLight™ 488	NHS, maleimide	493	518	1011	70,000	- Highly water-soluble and pH-insensitive
							FAM	SE	495	520		83,000	
							FITC		494	518	389	77,000	
							Fluorescein	NHS, maleimide	490	514	332	93,000	
CF™ 543	amine, hydrazide, maleimide, SE	541	560	870	100,000	532 nm or 543, 546 laser	Alexa Fluor® 546	N/A	556	573	N/A	112,000	- Significantly brighter than Alexa Fluor® 546 - Highly water-soluble and pH-insensitive
CF™ 555	amine, hydrazide, maleimide, SE	555	565	810	150,000	532 nm or 568 nm laser	Alexa Fluor® 555	maleimide, alkyne, azide, cadaverine, hydrazide, SE	555	572	1250	155,000	- Brighter and more photostable than Cy 3 - Minimal charge compared to Alexa Fluor® 555 reduces nonspecific antibody staining
							Cy 3	Bis NHS, hydrazide, maleimide, NHS	552	566	767	150,000	
							DY-547	NHS, maleimide, carboxylic acid, amino derivative	557	574	739	150,000	
							DyLight™ 550	NHS, maleimide	562	576	1040	150,000	
							TRITC		557	576	479	100,000	
CF™ 568	amine, aminoxy, hydrazide, maleimide, SE	562	583	714	100,000	532 nm or 568 nm laser	Alexa Fluor® 568	maleimide, cadaverine, hydrazide, SE	578	602	792	88,000	- Optimal for the 568 nm line of the Ar-Kr mixed-gas laser
							DY-560	NHS, maleimide, carboxylic acid, amino derivative	559	578	769	120,000	- Brighter and more photostable than Alexa Fluor® 568
							Rhodamine Red	NHS	570	590	654	129,000	
CF™ 594	amine, aminoxy, hydrazide, maleimide, SE	593	614	729	115,000	532 nm or 594 nm laser	Alexa Fluor® 594	maleimide, alkyne, azide, cadaverine, hydrazide, SE	590	617	820	92,000	- Yields the brightest antibody conjugates among spectrally similar dyes
							DY-594	NHS, maleimide	589	614		92,000	- Extremely photostable
							DyLight™ 594	NHS, maleimide	593	618	1078	80,000	
							Texas Red®	SE	595	615	702	116,000	
CF™ 620R	maleimide, SE	617	639	738	115,000	633 nm or 635 nm laser	LightCycler® Red 640	NHS	625	640	758		- Excellent energy acceptor for FRET - Highly fluorescent - Extremely photostable - Highly water-soluble
CF™ 633	amine, aminoxy, hydrazide, maleimide, SE	630	650	821	100,000	633 nm or 635 nm laser	Alexa Fluor® 633	maleimide, hydrazide, SE	621	639	1200	159,000	- Yields the brightest antibody conjugates among spectrally similar dyes Optimal for 633 nm He-Ne laser or the 635 nm red diode laser
							DY-630	NHS, maleimide, carboxylic acid, amino derivative	636	657	732	200,000	- Far more photostable than Alexa Fluor® 647
							DyLight™ 633	NHS, maleimide	638	658	1066	170,000	- Highly water-soluble
CF™ 640R	amine, aminoxy, hydrazide, maleimide, SE	642	662	832	105,000	633 nm, 635 nm, or 640 nm laser	Alexa Fluor® 647	maleimide, alkyne, azide, cadaverine, hydrazide, SE	651	672	1250	270,000	- Most photostable among spectrally similar dyes - Yields highly fluorescent protein conjugates
							Cy 5	Bis NHS, hydrazide, maleimide, NHS	649	666	792	250,000	- Very water-soluble and pH-insensitive

Fluorescent Dye Comparison cont'd

CF™ dye	Reactive forms	λEx (nm)	λEm (nm)	MW	ε	Excitation source	Direct replacement for:	Reactive forms	λEx (nm)	λEm (nm)	MW	ε	Advantages of CF™ dye
CF™ 647	hydrazide, maleimide, SE	650	665	836	240,000	633 nm, 635 nm, or 640 nm laser	Alexa Fluor® 647	maleimide, alkyne, azide, cadaverine, hydrazide, SE	651	672	1250	270,000	<ul style="list-style-type: none"> - Antibody conjugates have the best signal-to-noise ratio compared to Alexa Fluor® 647, DyLight™ 649 and Cy 5 - Highly fluorescent - Highly water-soluble and pH-insensitive
							Cy 5	Bis NHS, hydrazide, maleimide, NHS	649	666	792	250,000	
							Dy- 647	NHS, maleimide, carboxylic acid, amino derivative	653	672	762	250,000	
							DyLight™ 650	NHS, maleimide	652	672	1066	250,000	
CF™ 660C	maleimide, SE	667	685	3112	200,000	633 nm, 635 nm, or 640 nm laser	Alexa Fluor® 660	maleimide, SE	668	698	1100	132,000	<ul style="list-style-type: none"> - Much brighter than Alexa Fluor® 660 - More photostable than Alexa Fluor® 660 - Highly water-soluble
CF™ 660R	aminooxy, maleimide, SE	663	682	888	100,000	633 nm, 635 nm, or 640 nm laser	Alexa Fluor® 660	maleimide, SE	668	698	1100	132,000	<ul style="list-style-type: none"> - Brighter than Alexa Fluor® 660 - The most photostable 660 nm dye, ideal for confocal microscopy - Highly water-soluble
							Alexa Fluor® 680	maleimide, SE	684	707	1150	183,000	<ul style="list-style-type: none"> - The brightest among spectrally similar dyes
CF™ 680	maleimide, SE	681	698	3241	210,000	680 nm or 685 nm laser	Cy 5.5	Bis NHS, hydrazide, maleimide, NHS	675	695	1128	250,000	<ul style="list-style-type: none"> - Superior signal-to-noise ratio in immunostaining - Highly water-soluble and pH-insensitive
							DyLight™ 680	NHS, maleimide	692	712	950	140,000	
							IRDye® 680	NHS, maleimide	676	693	1402	250,000	
							Alexa Fluor® 680	maleimide, SE	684	707	1150	183,000	
CF™ 680R	aminooxy, maleimide, SE	680	701	912	140,000	680 nm or 685 nm laser	Cy 5.5	Bis NHS, hydrazide, maleimide, NHS	675	695	1128	250,000	<ul style="list-style-type: none"> - The most photostable 680 nm dye - Suitable for labeling nucleic acids and small bio-molecules - Highly water-soluble and pH-insensitive - CF680/CF770 double labeling compatible with Li-COR Odyssey System
							DyLight™ 680	NHS, maleimide	692	712	950	140,000	
							IRDye® 680	NHS, maleimide	676	693	1402	250,000	
							Alexa Fluor® 680	maleimide, SE	684	707	1150	183,000	
CF™ 750	SE	755	777	3009	250,000	680 nm, 685 nm, or 785 nm laser	Cy 7	Bis NHS, NHS	743	767	818	250,000	<ul style="list-style-type: none"> - Exceptionally bright and stable - Less immunogenic than competing dyes - Better signal-to-noise ratio compared to APC- Alexa Fluor® 750 tandem dye with 633 nm excitation
							DyLight™ 750	NHS, maleimide	752	778	1034	210,000	
							APC- Alexa Fluor® 750	Conjugated	650	779		240,000	
							DyLight™ 800	NHS, maleimide	777	794	899	270,000	
CF™ 770	SE	770	797	3138	220,000	785 nm laser	IRDye® 800CW	NHS, maleimide, carboxylate	778	794	1166	300,000	<ul style="list-style-type: none"> - Exceptionally bright and stable - Less immunogenic than competing dyes - CF680/CF770 double labeling compatible with Li-COR Odyssey System
CF™ 790	SE	784	806	3267	210,000	785 nm laser	Alexa Fluor® 790	SE	785	810	1750	260,000	<ul style="list-style-type: none"> - Exceptionally bright and stable - Less immunogenic than competing dyes

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