

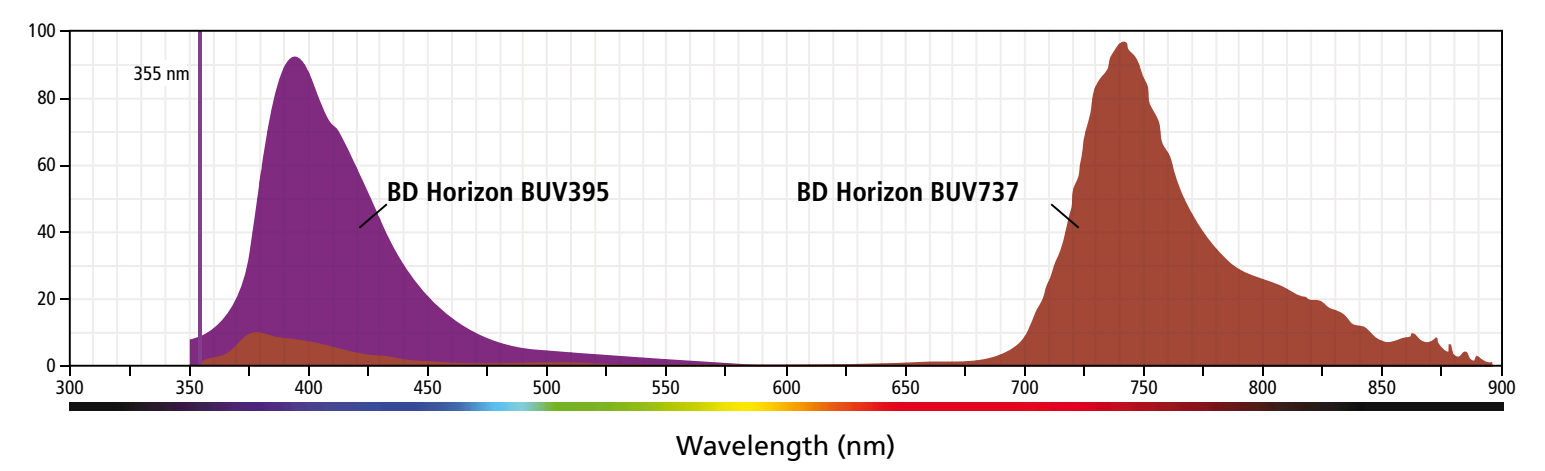
# BD Biosciences Fluorochrome/Laser Reference Poster

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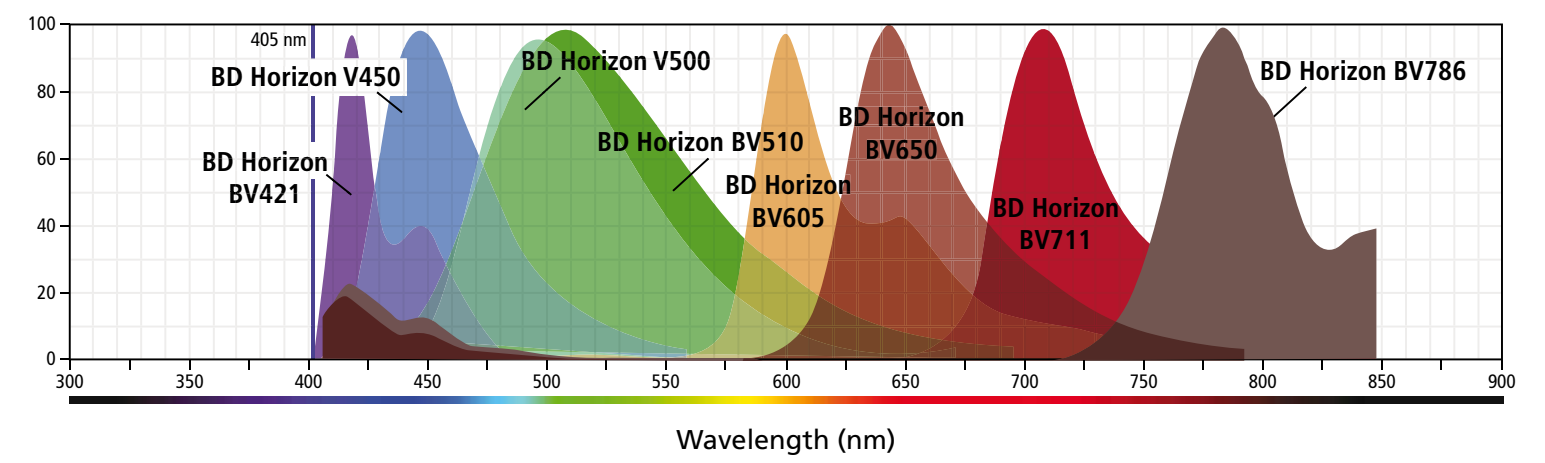
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Experience the full potential of multicolor flow cytometry with BD Biosciences flow cytometry instruments, reagents, and services. Visit our website for tools and information related to multicolor panel design including the interactive Fluorescence Spectrum Viewer, Multicolor Antibody Reagents Catalog, Human and Mouse Panels, and more.

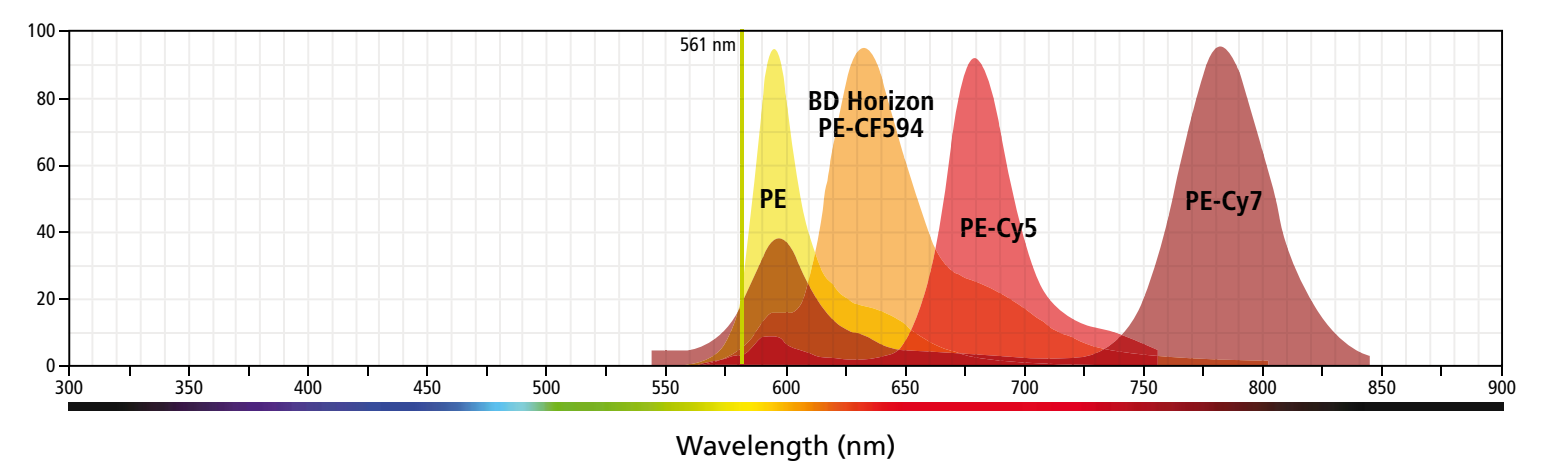
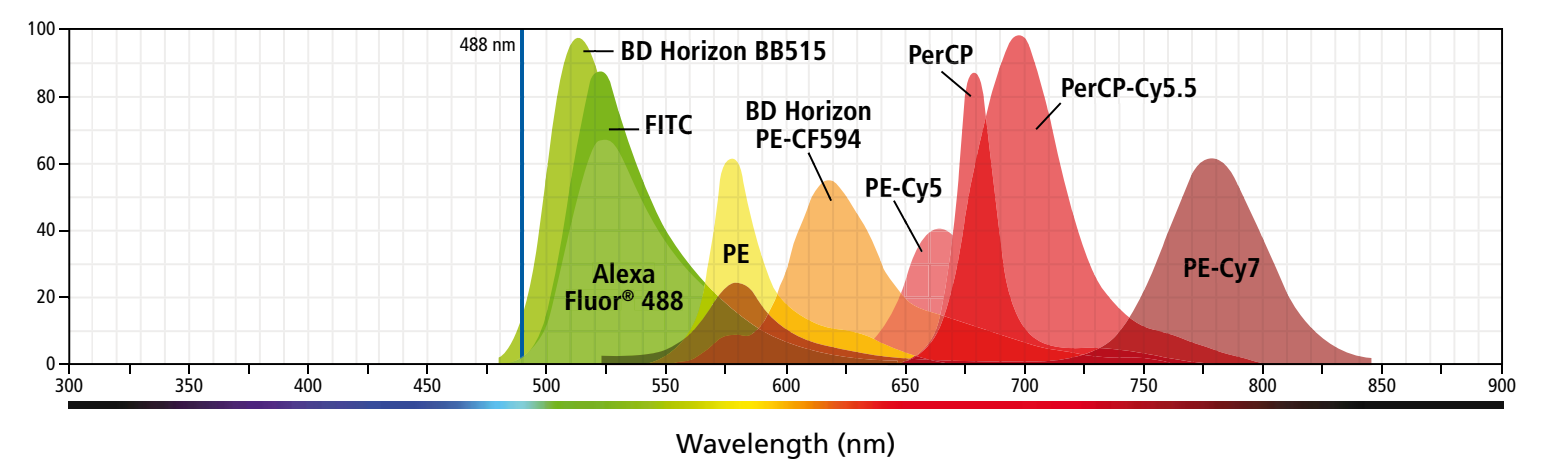
Analyzers				Sorters				Excitation Laser Line	Filter	Relative Brightness	Ultraviolet Laser (355 nm)
BD Accuri™ C6	BD FACSCalibur™	BD FACSVers™	BD FACSCanto™ II	BD LSRFortessa™ X-20	BD FACSAria™ Product Family	BD Influx™	BD FACStar™				
								355 nm	379/28	1	BD Horizon Brilliant™ Ultraviolet 395 (BUV395) (Ex <sub>max</sub> 348 nm/Em <sub>max</sub> 395 nm) is a UV-excitable dye that has been developed exclusively by BD Biosciences for instruments equipped with a 355-nm UV laser. This dye is optimal for multicolor flow cytometry because it has little to no spillover into other detectors.
								355 nm	740/35	2	BD Horizon Brilliant™ Ultraviolet 737 (BUV737) (Ex <sub>max</sub> 348 nm/Em <sub>max</sub> 737 nm) is a tandem fluorochrome that combines BD Horizon BUV395 and an acceptor dye with an Em Max at 737 nm. Due to the excitation of the acceptor dye by other laser lines, there may be significant spillover into channels detecting Alexa Fluor® 700 like dyes (eg. 712/20-nm filter). BUV737 has been exclusively developed by BD Biosciences for instruments equipped with a 355-nm UV laser.



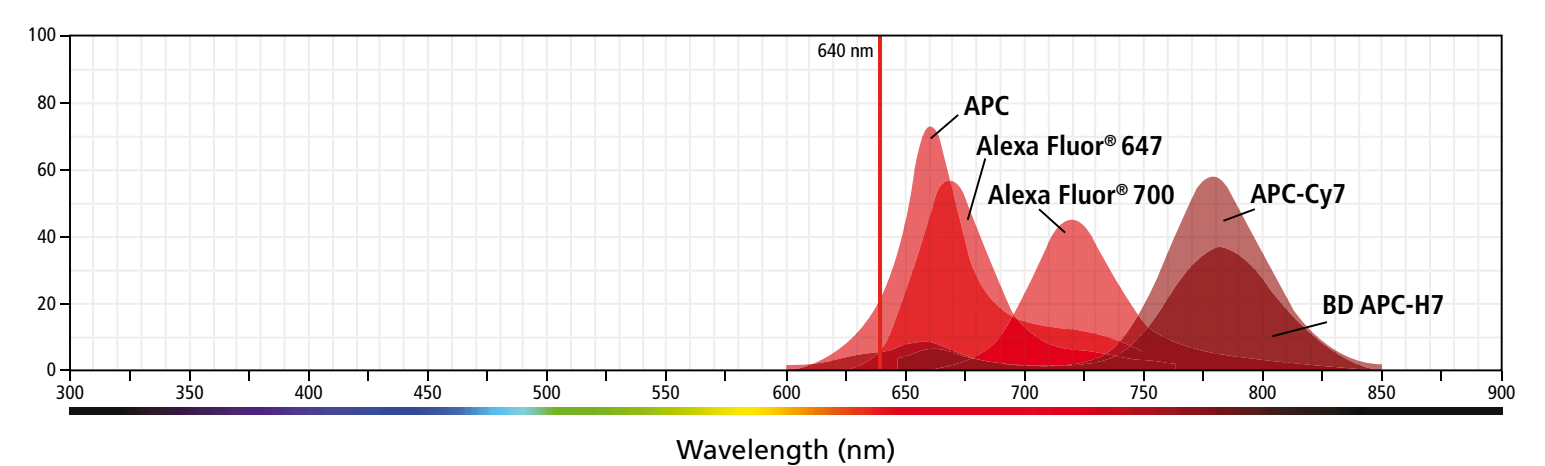
Analyzers				Sorters				Excitation Laser Line	Filter	Relative Brightness	Violet Laser (405 nm)
BD Accuri™ C6	BD FACSCalibur™	BD FACSVers™	BD FACSCanto™ II	BD LSRFortessa™ X-20	BD FACSAria™ Product Family	BD Influx™	BD FACStar™				
								405 nm	450/40	1	BD Horizon Brilliant™ Violet 421 (BV421) (Ex <sub>max</sub> 407 nm/Em <sub>max</sub> 421 nm) is a polymer-based dye excited by the violet laser and is one of the brightest fluorochromes offered by BD Biosciences. Conjugates are typically 10 times brighter than Pacific Blue™ conjugates and are often as bright as or brighter than PE conjugates. Due to similar excitation and emission properties, BD Horizon BV421 and BD Horizon V450 cannot be used simultaneously.
								405 nm	450/40	2	BD Horizon™ V450 (Ex <sub>max</sub> 404 nm/Em <sub>max</sub> 448 nm) is a coumarin dye excited by the violet laser. Due to similar excitation and emission properties but different spillover characteristics, BD Horizon BV421 and BD Horizon V450 cannot be used simultaneously.
								405 nm	525/50	3	BD Horizon Brilliant™ Violet 510 (BV510) (Ex <sub>max</sub> 405 nm/Em <sub>max</sub> 510 nm) is a polymer-based dye that is brighter than BD Horizon™ V500. Due to similar excitation and emission properties, BD Horizon BV510 and BD Horizon V500 cannot be used simultaneously.
								405 nm	525/50	4	BD Horizon™ V500 (Ex <sub>max</sub> 415 nm/Em <sub>max</sub> 500 nm) is a novel organic dye excited by the violet laser. This dye offers improved brightness over Pacific Orange™ and reduced spillover into the FITC channel when compared to AmCyan. BD Horizon V500 cannot be used simultaneously with BD Horizon BV510 or Pacific Orange™.
								405 nm	610/20	5	BD Horizon Brilliant™ Violet 605 (BV605) (Ex <sub>max</sub> 407 nm/Em <sub>max</sub> 602 nm) is a tandem fluorochrome that combines BD Horizon BV421 and Cy™3.5. These conjugates are very bright, exhibiting similar brightness to equivalent PE conjugates. Due to the excitation of Cy3.5 by the green (532-nm) and yellow-green (561-nm) lasers, there will be significant spillover of the BD Horizon BV605 signal into the PE and BD Horizon PE-CF594 detectors off the green or yellow-green lasers.
								405 nm	660/20	6	BD Horizon Brilliant™ Violet 650 (BV650) (Ex <sub>max</sub> 407 nm/Em <sub>max</sub> 650 nm) is a tandem fluorochrome of BD Horizon™ BV421 and an acceptor dye with an Em Max at 650 nm. Due to the excitation and emission characteristics of the acceptor dye, there will be spillover into the APC and Alexa Fluor® 700 detectors. BD Horizon BV650 will have moderate spillover into the BD Horizon™ BV711 detector.
								405 nm	710/50	7	BD Horizon Brilliant™ Violet 711 (BV711) (Ex <sub>max</sub> 407 nm/Em <sub>max</sub> 711 nm) is a tandem fluorochrome of BD Horizon BV421 and an acceptor dye with an Em Max at 711 nm. This dye offers a very bright choice for the violet laser. Due to the excitation and emission characteristics of the acceptor dye, there may be moderate spillover into the Alexa Fluor® 700 and PerCP-Cy™5.5 detectors. BD Horizon BV711 will also have moderate spillover into the BD Horizon™ BV786 detector.
								405 nm	780/60	8	BD Horizon Brilliant™ Violet 786 (BV786) (Ex <sub>max</sub> 407 nm/Em <sub>max</sub> 786 nm) is a tandem fluorochrome of BD Horizon BV421 and an acceptor dye with an Em Max at 786 nm. BD Horizon BV786 offers a bright choice for the sixth detector off the violet laser.



Analyzers				Sorters				Excitation Laser Line	Filter	Relative Brightness	Blue Laser (488 nm) / Yellow-Green Laser (561 nm)
BD Accuri™ C6	BD FACSCalibur™	BD FACSVers™	BD FACSCanto™ II	BD LSRFortessa™ X-20	BD FACSAria™ Product Family	BD Influx™	BD FACStar™				
								488 nm	530/30	1	BD Horizon Brilliant™ Blue 515 (BB515) (Ex <sub>max</sub> 490 nm/Em <sub>max</sub> 515 nm) is a dye that was exclusively developed by BD Biosciences as a brighter alternative to FITC. This dye is up to seven times brighter than FITC and has less spillover into the PE channel. Due to similar excitation and emission properties, BD Horizon BB515 and FITC/Alexa Fluor® 488 cannot be used simultaneously.
								488 nm	530/30	2	Alexa Fluor® 488 (Ex <sub>max</sub> 495 nm/Em <sub>max</sub> 519 nm) conjugates are highly photostable and remain fluorescent over a broad pH range. Alexa Fluor® 488 tends to be brighter than FITC and more optimal for intracellular applications. Due to nearly identical excitation and emission properties, FITC and Alexa Fluor® 488 cannot be used simultaneously. Alexa Fluor® 488 exhibits extraordinary photostability, which makes it highly suitable for fluorescence microscopy.
								488 nm	530/30	3	FITC (Ex <sub>max</sub> 494 nm/Em <sub>max</sub> 520 nm) Fluorescein isothiocyanate (FITC) is a fluorochrome with a molecular weight of 389 Da. FITC is sensitive to pH changes and photobleaching. Due to nearly identical excitation and emission properties, FITC and Alexa Fluor® 488 cannot be used simultaneously. FITC is relatively dim and should be reserved for highly expressed markers whenever possible.
								488 nm 532 nm 561 nm	575/26	4	PE (Ex <sub>max</sub> 496 nm/Em <sub>max</sub> 578 nm) R-phycoerythrin (PE) is an accessory photosynthetic pigment found in red algae. It exists in vitro as a 240-kDa protein with 23 phycoerythrin chromophores per molecule. This makes PE the brightest fluorochrome for flow cytometry applications, but its photobleaching properties make it unsuitable for fluorescence microscopy.
								488 nm 532 nm 561 nm	610/20	5	BD Horizon™ PE-CF594 (Ex <sub>max</sub> 496 nm/Em <sub>max</sub> 612 nm) is a tandem conjugate, developed exclusively by BD Biosciences, that combines PE and CF594. PE-CF594 is a brighter alternative to PE-Texas Red® with improved spectral characteristics.
								488 nm 532 nm 561 nm	670/14	6	PE-Cy™5 (Ex <sub>max</sub> 496 nm/Em <sub>max</sub> 667 nm) is a tandem conjugate that combines phycoerythrin and the cyanine dye Cy5. Because of its broad absorption range and the fact that its emission spectra are equivalent to APC, PE-Cy5 is not recommended for simultaneous use with APC. The Cy5 molecule has been shown to exhibit nonspecific binding to Fc receptors, which is most apparent on monocyte populations.
								488 nm 532 nm	695/40	7	PerCP (Ex <sub>max</sub> 482 nm/Em <sub>max</sub> 678 nm) is a component of the photosynthetic apparatus found in the dinoflagellate <i>Glenodinium</i> . PerCP is a protein complex with a molecular weight of ~35 kDa. Due to its photobleaching characteristics, PerCP conjugates are not recommended for use on flow cytometers with high-power lasers (>25 mW).
								488 nm 532 nm	695/40	8	PerCP-Cy™5.5 (Ex <sub>max</sub> 482 nm/Em <sub>max</sub> 695 nm) is a tandem conjugate that combines PerCP with the cyanine dye Cy5.5. PerCP-Cy5.5 is not subject to photobleaching like PerCP and can be used with stream-in-air flow cytometers. Additionally, the PerCP-Cy5.5 tandem conjugate is not as susceptible to fixative or light instability compared to APC-Cy™7 and PE-Cy7.
								488 nm 532 nm 561 nm	780/60	9	PE-Cy™7 (Ex <sub>max</sub> 496 nm/Em <sub>max</sub> 785 nm) is a tandem fluorochrome that combines PE and the cyanine dye Cy7. PE-Cy7 is sensitive to photo-induced degradation, resulting in loss of fluorescence and changes in spillover. Extreme caution must be taken to avoid light exposure and prolonged exposure to paraformaldehyde fixative. Fixed cells should be analyzed within 4 hours of fixation in paraformaldehyde or transferred to a paraformaldehyde-free buffer for overnight storage.



Analyzers				Sorters				Excitation Laser Line	Filter	Relative Brightness	Red Laser (640 nm)
BD Accuri™ C6	BD FACSCalibur™	BD FACSVers™	BD FACSCanto™ II	BD LSRFortessa™ X-20	BD FACSAria™ Product Family	BD Influx™	BD FACStar™				
								633 nm 635 nm 640 nm	660/20	1	APC (Ex <sub>max</sub> 650 nm/Em <sub>max</sub> 660 nm), Allophycocyanin (APC), is an accessory photosynthetic pigment found in blue-green algae. Its molecular weight is approximately 105 kDa. Due to nearly identical excitation and emission properties, APC and Alexa Fluor® 647 cannot be used simultaneously.
								633 nm 635 nm 640 nm	660/20	2	Alexa Fluor® 647 (Ex <sub>max</sub> 650 nm/Em <sub>max</sub> 668 nm) conjugates are highly photostable and remain fluorescent over a broad pH range. Due to nearly identical excitation and emission properties, APC and Alexa Fluor® 647 cannot be used simultaneously. APC tends to be brighter while Alexa Fluor® 647 is more optimal for intracellular applications. This fluorochrome exhibits uncommon photostability, making it an ideal choice for use in fluorescence microscopy.
								633 nm 635 nm 640 nm	730/45	3	Alexa Fluor® 700 (Ex <sub>max</sub> 696 nm/Em <sub>max</sub> 719 nm) is a far-red dye that can be excited with a 633-640-nm laser. This enables multicolor analysis in conjunction with APC or Alexa Fluor® 647 and APC-H7 or APC-Cy7 reagents.
								633 nm 635 nm 640 nm	780/60	4	APC-Cy™7 (Ex <sub>max</sub> 650 nm/Em <sub>max</sub> 785 nm) is a tandem fluorochrome that combines APC and the cyanine dye Cy7. Special precautions must be taken with APC-Cy7 conjugates, and cells stained with them, to protect the fluorochrome from long-term exposure to light. Fixed cells should be analyzed within 4 hours of fixation in paraformaldehyde or transferred to a paraformaldehyde-free buffer for overnight storage. Due to nearly identical excitation and emission properties, APC-Cy7 and APC-H7 cannot be used simultaneously.
								633 nm 635 nm 640 nm	780/60	5	APC-H7 (Ex <sub>max</sub> 650 nm/Em <sub>max</sub> 785 nm) is an APC-cyanine tandem fluorochrome which uses an analog of Cy7 and has similar spectral properties to APC-Cy7. APC-H7 conjugates provide greater stability in light and paraformaldehyde fixatives and have less spillover into the APC channel than APC-Cy7 conjugates. Due to nearly identical excitation and emission properties, APC-Cy7 and APC-H7 cannot be used simultaneously.



Relative Brightness Key: Dim Moderate Bright Brightest

Brightest dyes will be about as bright as PE while Dim dyes will have brightness similar to BD Horizon V500. Relative brightness is dependent on instrument configuration including lasers, filters, and laser power.

\* Capable of detecting 8 colors simultaneously (4 blue laser, 2 red laser, 2 violet laser). PE-CF594 and Alexa Fluor® 700 filters are available separately.

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APC-Cy7: US Patent 5,714,386

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