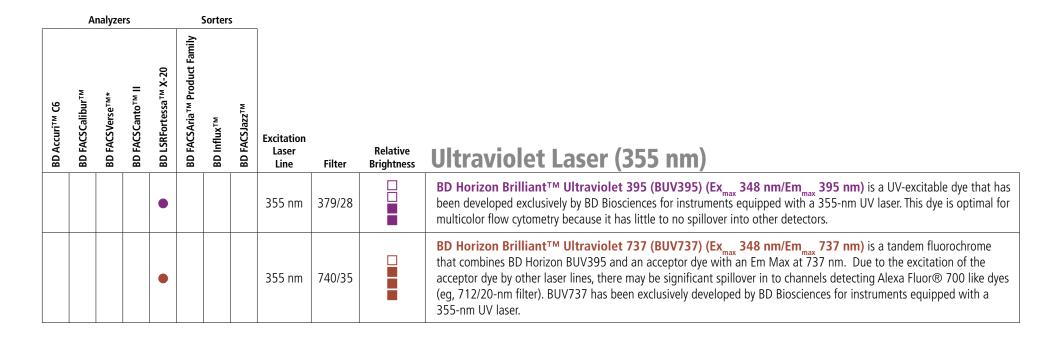
BD Biosciences

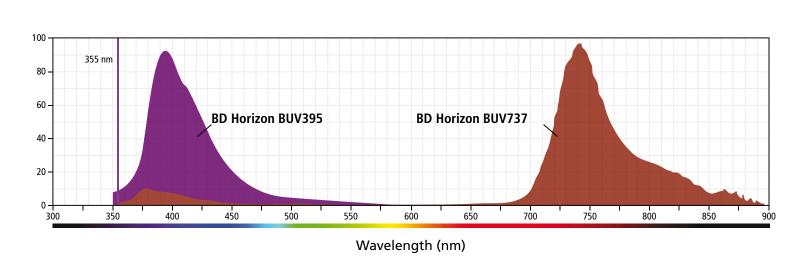
Fluorochrome/Laser Reference Poster

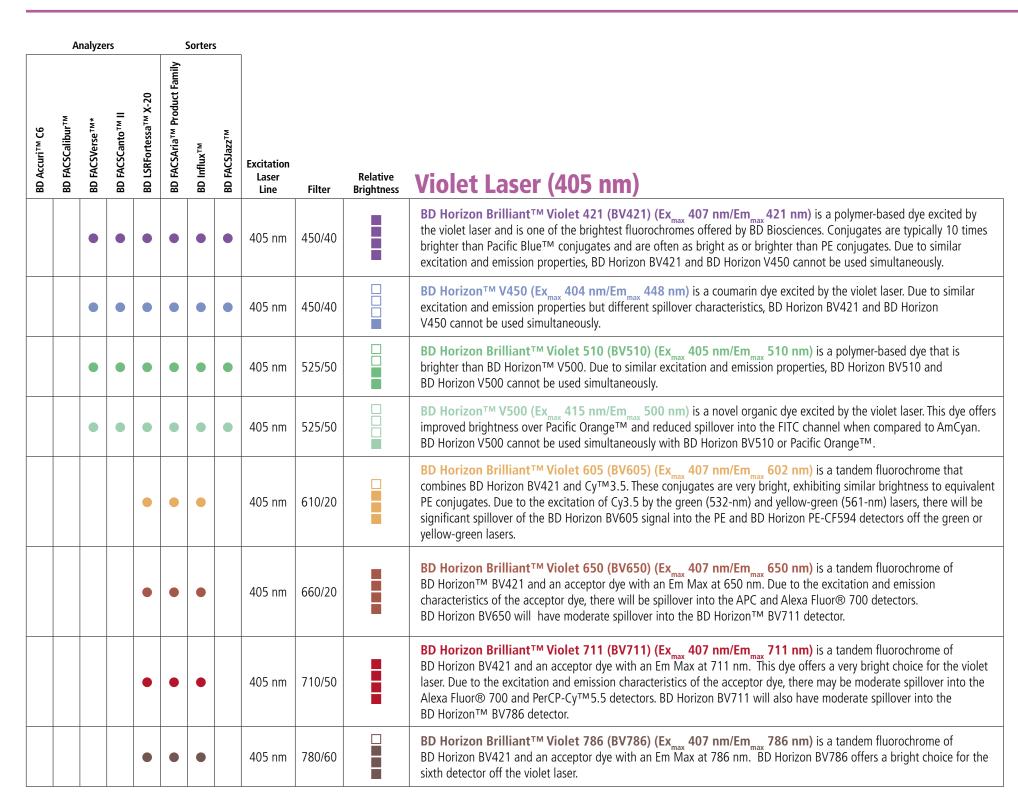
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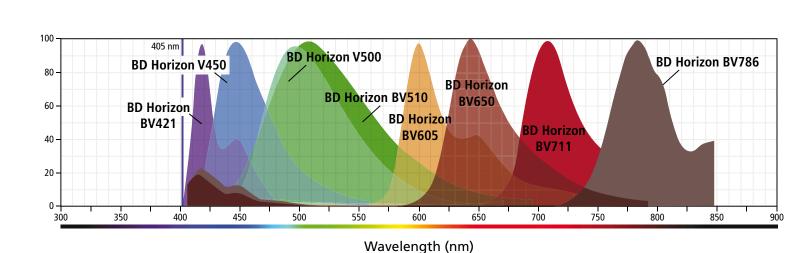
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Experience the full potential of multicolor flow cytometry with BD Biosciences flow cytometry instruments, reagents, and services. Visit our website for tools and information related to multicolor panel design including the interactive Fluorescence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

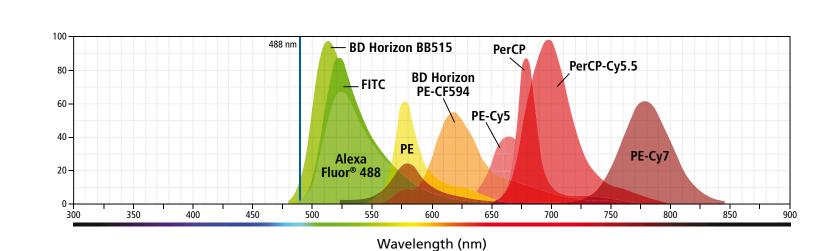


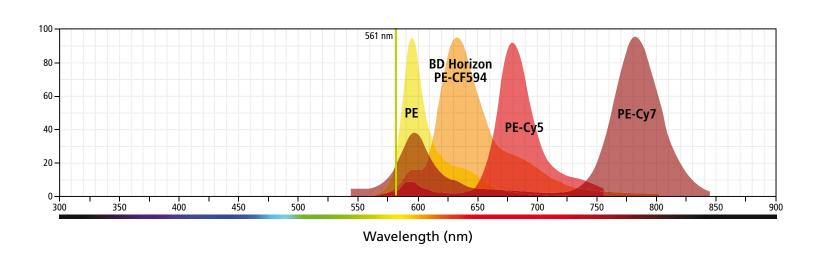






Analyzers Sorters Excitation Laser Blue Laser (488 nm) / Yellow-Green Laser (561 nm) BD Horizon Brilliant™ Blue 515 (BB515) (Ex_{max} 490 nm/Em_{max} 515 nm) is a dye that was exclusively developed by BD Biosciences as a brighter alternative to FITC. This dye is up to seven times brighter than FITC and 530/30 488 nm 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. Alexa Fluor® 488 (Ex., 495 nm/Em, 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 488 nm 530/30 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. FITC (Ex 494 nm/Em 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, 530/30 FITC and Alexa Fluor® 488 cannot be used simultaneously. FITC is relatively dim and should be reserved for highly expressed markers whenever possible. PE (Ex., 496 nm/Em., 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 phycoerythrobilin chromophores per molecule. This makes PE the brightest 575/26 532 nm fluorochrome for flow cytometry applications, but its photobleaching properties make it unsuitable for fluorescence 561 nm BD Horizon™ PE-CF594 (Ex., 496 nm/Em., 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 532 nm 610/20 561 nm spectral characteristics. PE-Cy[™]5 (Ex_{max}496 nm/Em_{max}667 nm) is a tandem conjugate that combines phycoerythrin and the cyanine dye 488 nm Cy5. Because of its broad absorption range and the fact that its emission spectra are equivalent to APC, PE-Cy5 is not 532 nm 670/14 recommended for simultaneous use with APC. The Cy5 molecule has been shown to exhibit nonspecific binding to Fc 561 nm receptors, which is most apparent on monocyte populations. PerCP (Ex., 482 nm/Em., 678 nm) is a component of the photosynthetic apparatus found in the dinoflagellate 488 nm 695/40 Glenodinium. PerCP is a protein complex with a molecular weight of ~35 kDa. Due to its photobleaching characteristics, 532 nm PerCP conjugates are not recommended for use on flow cytometers with high-power lasers (>25 mW). PerCP-Cy[™]5.5 (Ex. 482 nm/Em. 695 nm) is a tandem conjugate that combines PerCP with the cyanine dye Cy5.5. PerCP-Cy5.5 is not subject to photobeaching like PerCP and can be used with stream-in-air flow cytometers. 695/40 532 nm Additionally, the PerCP-Cy5.5 tandem conjugate is not as susceptible to fixative or light instability compared to APC-Cy™7 and PE-Cy7. PE-Cy[™]7 (Ex., 496 nm/Em., 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 488 nm





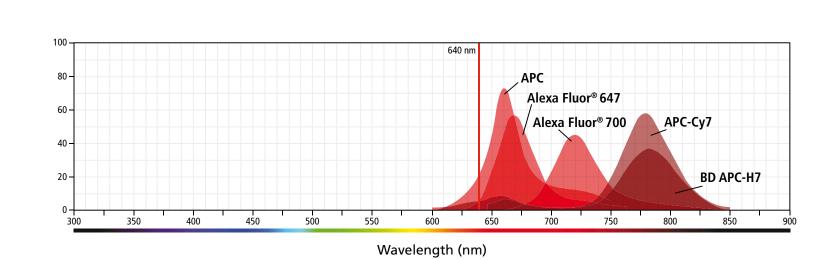
Analyzers						Sorter	s				
BD Accuri™ C6	BD FACSCalibur™	BD FACSVerse™*	BD FACSCanto™ II	BD LSRFortessa™ X-20	BD FACSAria™ Product Family	BD Influx™	BD FACSJazz™	Excitation Laser Line	Filter	Relative Brightness	Red Laser (640 nm)
•	•			•	•	•	•	633 nm 635 nm 640 nm	660/20		APC (Ex _{max} 650 nm/Em _{max} 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		Alexa Fluor® 647 (Ex _{max} 650 nm/Em _{max} 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		Alexa Fluor® 700 (Ex _{max} 696 nm/Em _{max} 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		APC-Cy TM 7 (Ex _{max} 650 nm/Em _{max} 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		APC-H7 (Ex _{max} 650 nm/Em _{max} 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.

overnight storage.

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





640 nm

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.

780/60

532 nm

561 nm

For Research Use Only. Not for use in diagnostic or therapeutic procedures. APC-Cy7: US patent 5,714,386

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