BD FACSCelesta™ Flow Cytometer Configuration Sheet
Blue-Violet-Red (BVR) Laser Configuration

The BD FACSCelesta™ flow cytometer is designed to simplify the use of multicolor flow cytometry and allow researchers to benefit from new innovations in instrument and reagent technology. This platform offers multiple configurations to meet varied application needs.

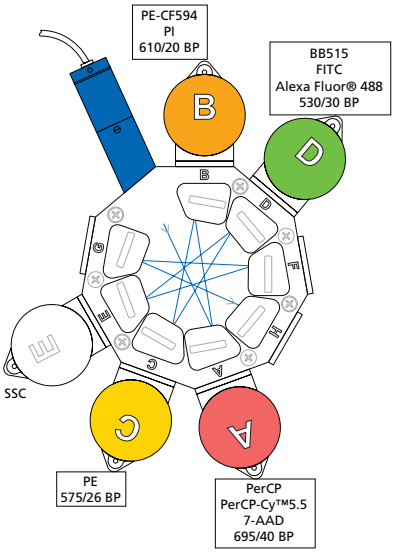
The BVR configuration of the BD FACSCelesta includes blue, violet, and red lasers, optimized to get the most out of the legacy fluorochromes, viability dyes, and BD Horizon Brilliant™ dyes shown in the optical path diagrams. While a blue laser has long been standard in flow cytometry, the violet laser is gaining popularity as more bright violet-excited fluorochromes, such as BD Horizon Brilliant™ Violet reagents, are introduced and enthusiastically adopted. The red laser, which optimally excites allophycocyanin (APC) and APC-based tandem dyes, allows markers to be spread across more lasers, further reducing the overall compensation needed for a multicolor panel.

Multicolor flow cytometry panel design has presented challenges for researchers, such as varying marker expression, varying dye brightness, and significant emission spillover between fluorescence channels. The combination of bright, narrow-spectrum, BD Horizon Brilliant fluorochromes and sensitive optics results in panels that can readily resolve even dim populations, yet are easy to use and compensate. The system operates with BD FACSDiva™ software, a collection of convenient and easy-to-use tools for flow cytometer and application setup, data acquisition, and data analysis.

Optical path diagrams and fluorochrome support

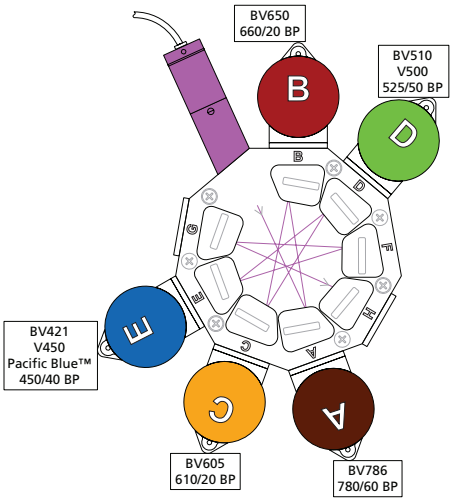
Laser polygons show fluorochromes, mirrors, filters, and optical paths for the BD FACSCelesta BVR configuration.

Blue laser



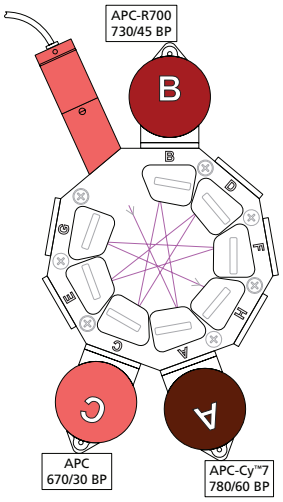
Other fluorochromes supported:
BD Horizon™ Fixable Viability Stain 520, 570, 620

Violet laser

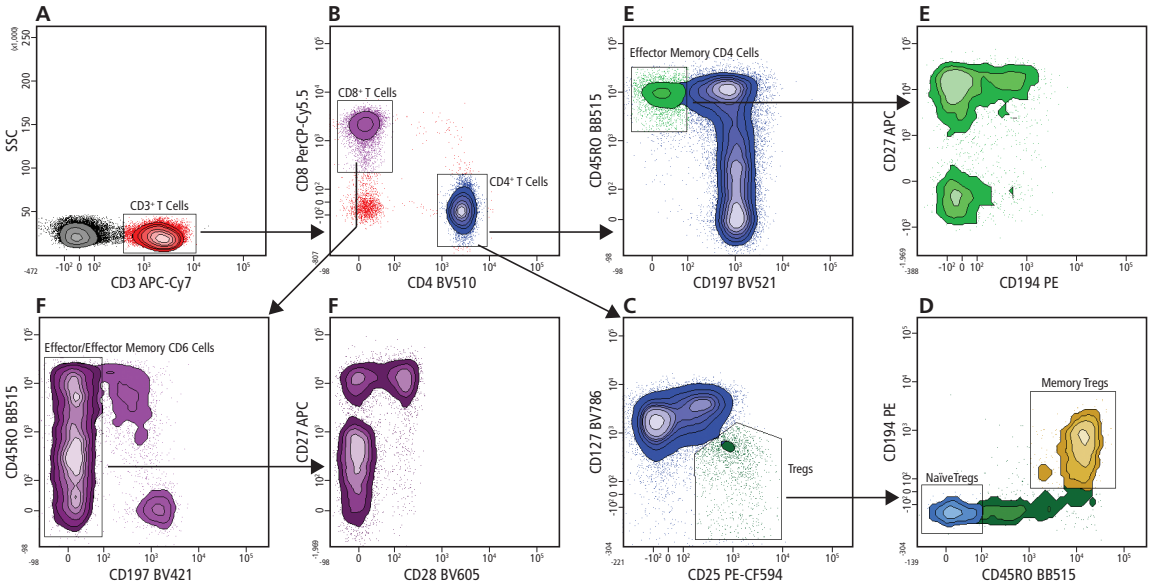


Other fluorochromes supported:
BD Horizon™ Fixable Viability Stain 450, 510

Red laser

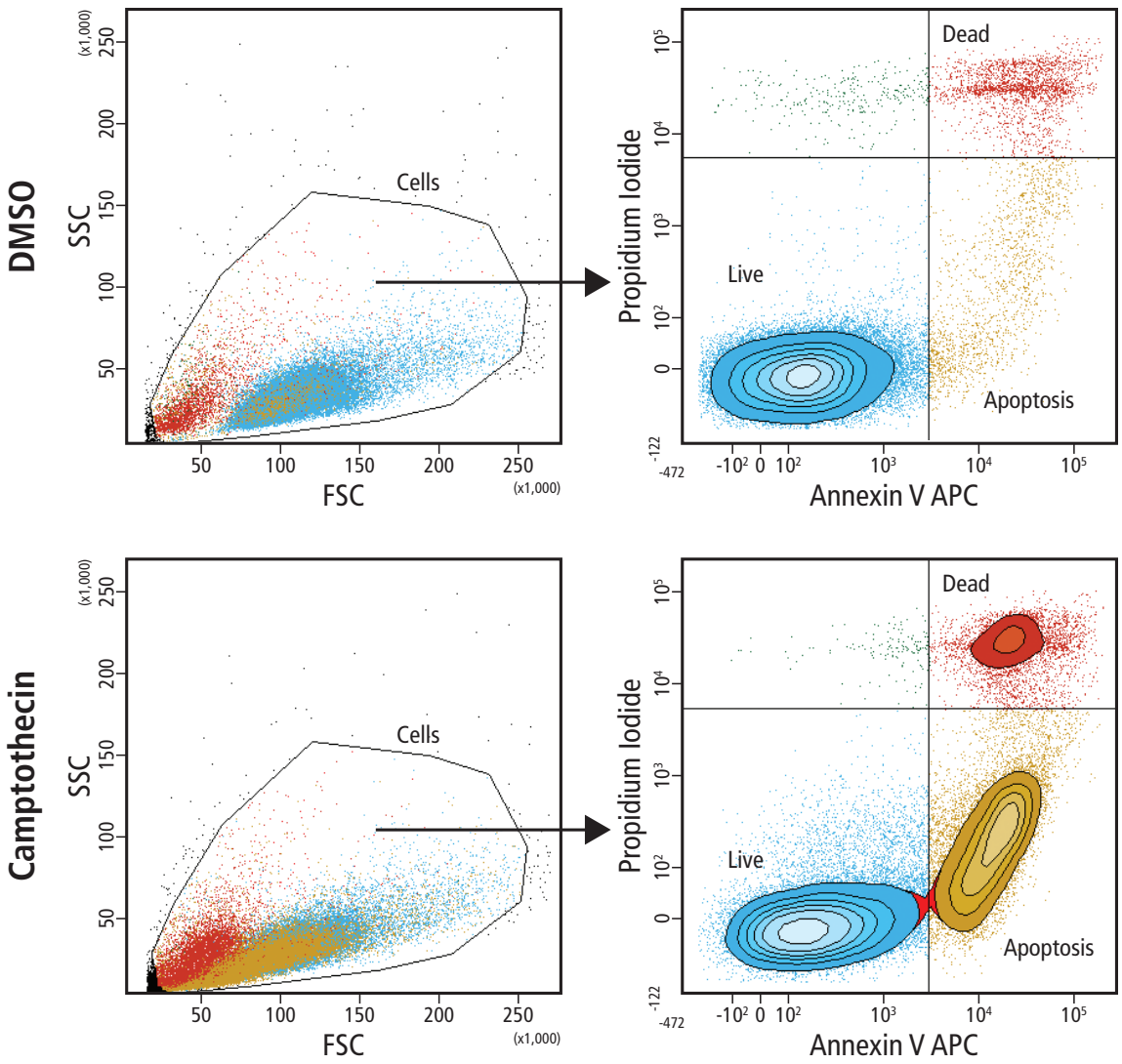


Other fluorochromes supported:
BD Horizon™ Fixable Viability Stain 660, 700, 780



10-color T-cell analysis on the BD FACSCelesta BVR configuration

This T-cell panel demonstrates the sensitivity and resolution of the BD FACSCelesta system, even in detecting rare subpopulations. After normal human whole blood was washed and lysed, BD Horizon Brilliant and traditional dyes were used to identify rare T-cell and Treg subpopulations. **A.** Cells were gated to select the $CD3^+$ T cells. **B.** $CD3^+$ lymphocytes were gated to show the $CD4^+$ helper T cells and $CD8^+$ cytotoxic T cells. **C.** Gated on the $CD4^+$ T cells, surface markers were used to identify $CD25^+CD127^-$ Tregs. **D.** Gated on Tregs, surface markers were used to identify memory and naïve Treg subsets. **E.** $CD4^+$ helper T cells were analyzed for memory T-cell subsets using CD45RO and CD197 (Left) and Effector Memory subsets were further defined with surface markers for CD27 and CD194 (Right). **F.** CD8+ cytotoxic T cells were analyzed for Effector and memory subsets using CD45RO and CD197 (Left) and Effector/effector memory subsets were further defined using CD27 and CD28 surface markers. Similar analysis was completed for $CD8^+$ cytotoxic T cells (Right).



Two-color flow cytometric analysis of apoptosis and viability in Jurkat cells

Jurkat cells were treated with 0.025% DMSO vehicle (top plots) or 5 µM camptothecin (bottom plots) for 4 hours, harvested from culture, washed, and resuspended in Annexin V Binding Buffer. Cells were then incubated with BD Pharmingen™ APC Annexin V (Cat. No. 550475) and BD Pharmingen™ Propidium Iodide Staining Solution (Cat. No. 556463) (PI) for 15 minutes at room temperature protected from light, and then analyzed by flow cytometry on a BD FACSCelesta system. Debris was excluded based on the light scatter properties of Jurkat cells (left plots). DMSO vehicle-treated cells were primarily Annexin V^-PI^-, indicating that most cells were live. Camptothecin-treated cells show an increase in the number of Annexin V^+ (apoptotic) and Annexin V^+PI^+ (dead) cells, indicating that camptothecin treatment induced apoptosis and cell death.

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Excitation Laser	Fluorochrome	Ex_{max}	Em_{max}	Relative Brightness
Violet (405 nm)	BD Horizon Brilliant™ Violet 786 (BV786)	407 nm	786 nm	
	BD Horizon Brilliant™ Violet 650 (BV650)	407 nm	650 nm	
	BD Horizon Brilliant™ Violet 605 (BV605)	407 nm	602 nm	
	BD Horizon Brilliant™ Violet 510 (BV510)	405 nm	510 nm	
	BD Horizon Brilliant™ Violet 421 (BV421)	407 nm	421 nm	
Blue (488 nm)	PerCP	482 nm	678 nm	
	PerCP-Cy™5.5	482 nm	695 nm	
	BD Horizon™ PE-CF594	496 nm	612 nm	
	PE	496 nm	578 nm	
	BD Horizon Brilliant™ Blue 515 (BB515)	490 nm	515 nm	
	FITC	494 nm	520 nm	
	Alexa Fluor® 488	495 nm	519 nm	
Red (640 nm)	APC-Cy™7	650 nm	785 nm	
	APC-R700	652 nm	704 nm	
	APC	660 nm	660 nm	

Relative Brightness Key:

- Dim
- Moderate
- Bright
- Brightest

Ordering Information

Description	Cat. No.
BD FACSCelesta™ Flow Cytometer, BVR Configuration	**660344**
BD FACSCelesta™ High Throughput Sampler (HTS) Option	**658946**
BD FACSFlow™ Supply System	**649908**
BD FACSCelesta™ Standard Workstation Bundle	**660472**

23-17600-00