BD LSRFortessa™

BD

Helping all people live healthy lives

Performance without peer,
choice without compromise

Performance without peer, choice without compromise

The BD LSRFortessa™ cell analyzer offers the ultimate in choice for flow cytometry, providing power, performance, and consistency. Designed to be affordable and expandable, the BD LSRFortessa has the flexibility to support the expanding needs of multicolor flow cytometry assays.

The BD LSRFortessa analyzer can be configured with up to 4 lasers—blue, red, violet, and UV—which enables the detection of up to 18 colors simultaneously. The BD LSRFortessa may be upgraded subsequently with additional or new lasers from BD, as future requirements dictate.

Innovative Designs for Optical Efficiency
Built on a proven platform, BD LSR analyzers feature innovative designs for both the excitation optics and collection optics that reduce excitation losses and improve light collection efficiency. The result is optical efficiency that delivers maximum sensitivity and resolution for multicolor applications by reducing excitation laser light loss, and improved fluorescent signal collection.

Special Order Program
For research needs at the leading edge of biomedical discovery, the special order program offers BD LSRFortessa researchers the latest innovations in laser technology to enable future upgrades with new excitation, wavelength, and power choices. The program lets customers fully customize configurations to deliver added flexibility and power to support advanced research.

Technical and Application Support
As with all BD instruments and reagents, highly qualified BD technical and application support personnel are available for help in streamlining research and maintaining optimal instrument performance.

Boundless Potential

The BD LSRFortessa Analyzer

The BD LSRFortessa is designed to enable your current assay requirements. As future needs arise, lasers can be added or upgraded.

BD High Throughput Sampler (HTS)

Flexibility in a Small Space

An affordable choice to fit most flow cytometry analyzer needs, the BD LSRFortessa can be ordered with up to 4 lasers—blue, red, violet, and UV—which provides flexibility in laser wavelengths so assay design can be optimized using the latest fluorescent dyes and substrates. The instrument can accommodate the detection of up to 18 colors simultaneously with a defined set of optical filters that meet or exceed the majority of today's assay requirements.

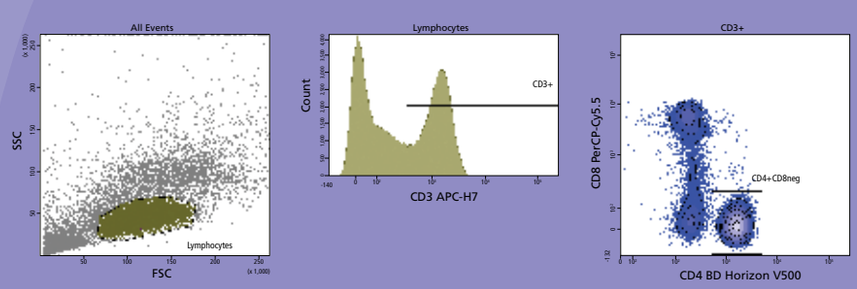
The BD LSRFortessa puts the power of the proven BD LSR platform into a compact footprint. It can easily fit on the benchtop for cost-effective space use. This space efficiency is even more important for labs with limited space or where space cost is premium.

In addition to the reduced size, design enhancements provide easier access to bandpass filters and mirrors, simplifying changes to experimental setup.

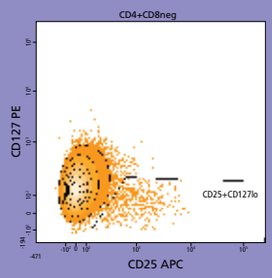
To improve experimental workflow, the optional BD™ High Throughput Sampler (HTS) provides rapid, fully automated sample acquisition from microtiter plates. In high-throughput mode, the HTS option can speed through a 96-well plate in less than 15 minutes with less than 0.5% sample carryover from one well to the next. Low carryover is essential in research applications to ensure sample purity and data integrity.

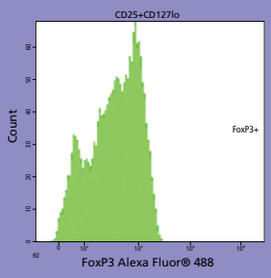
Analysis of Regulatory T Cells

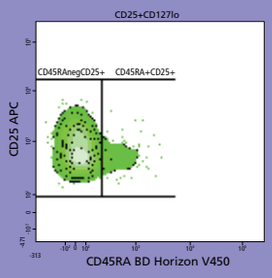
Regulatory T cells (Tregs) play a critical role in maintaining immune modulation and are present in normal peripheral blood in low numbers (5 to 10% of $CD4^+$ T cells or 1 to 2% of total lymphocytes). Forkhead box P3 (FoxP3) is considered to be the definitive marker for this rare cell population. Tregs are known to express the highest levels of CD25. The $CD4^+CD25^{+(high)}$ gating strategy shown here was used to identify Treg populations. These plots represent data taken from a BD LSRFortessa using an 8-color panel that applies this gating strategy to identify FoxP3-positive cells.

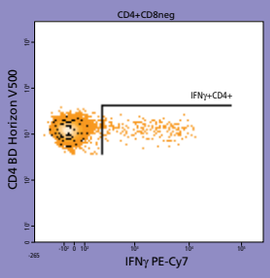


BD LSRFORTESSA









Tube: Stimulated

Population	#Events	%Parent	%Total
All Events	960,606	####	100.0
Lymphocytes	238,144	24.8	24.8
CD3+	108,466	45.5	11.3
CD4+CD8neg	63,347	58.4	6.6
CD25+CD127lo	4,059	6.4	0.4
FoxP3+	3,192	78.6	0.3
CD45RA+CD25+	3,360	82.8	0.3
CD45RAnegCD25+	699	17.2	0.1
IFNg+CD4+	1,921	3.0	0.2

Superior Performance

Unique and Revolutionary Designs for Multicolor Analysis

Many innovations are incorporated into the BD LSRFortessa product line. The heart of the cytometer, the fluidics system features a true fixed-alignment flow cell that is gel-coupled to the collection optics to maximize detector signal.

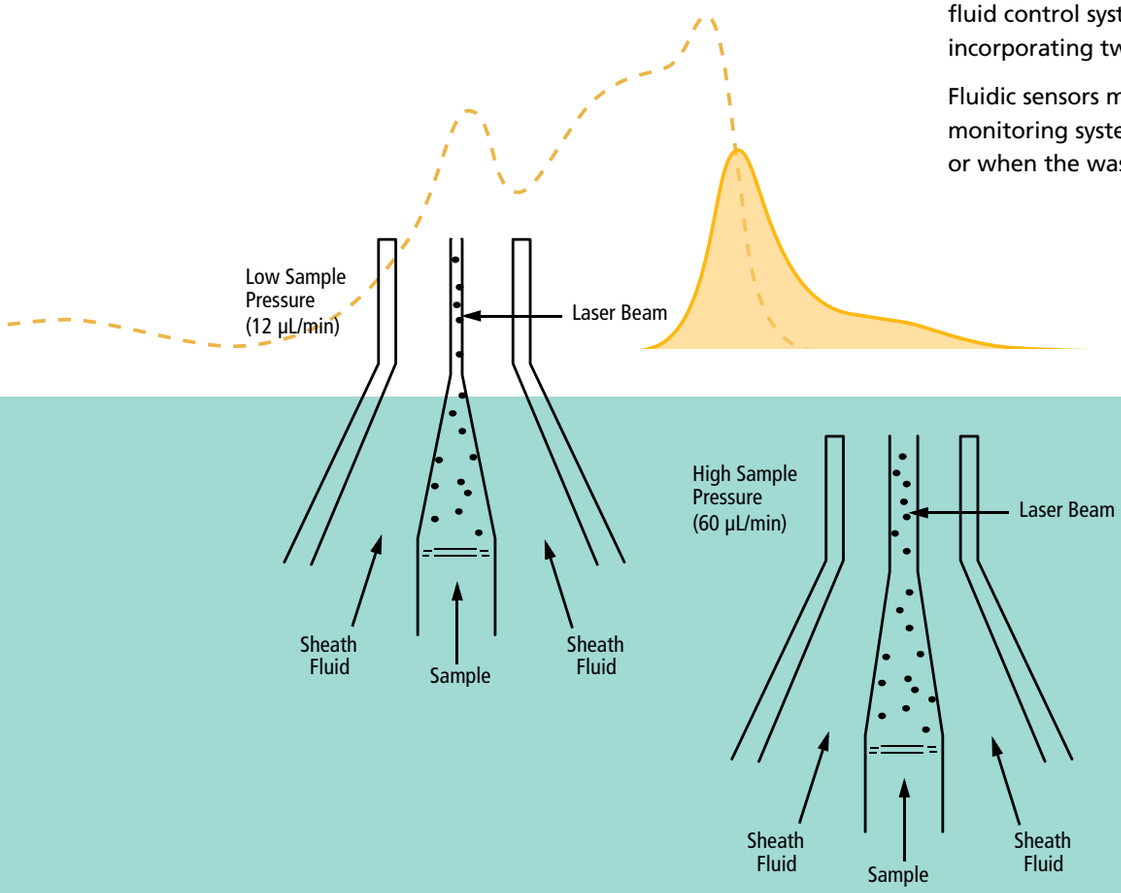
Fluidics System

The fluidics system is pressure driven. Hydrodynamic focusing forces sample cells through the cuvette flow cell, where they are interrogated. The flow cell is in fixed alignment with the laser and gel-coupled to the collection optics. This design ensures that the laser is precisely focused on the sample stream and the maximum amount of emitted light can be collected for added sensitivity in multicolor applications. Fixed alignment also minimizes startup time, improves experiment-to-experiment reproducibility, and enables automated daily quality control.

The sheath container (8 L) and waste container (10 L) are outside the cytometer, and positioned on the floor for easier access.

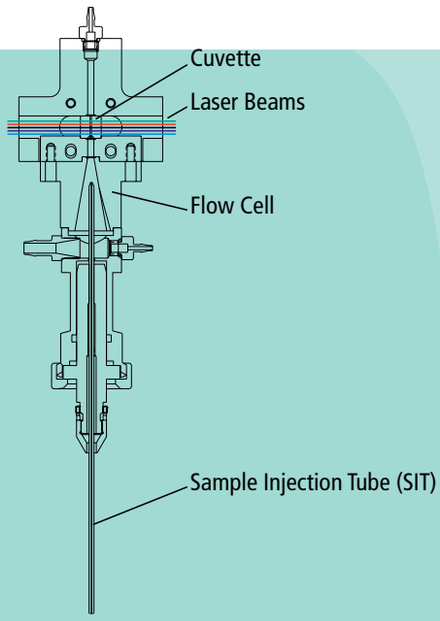
The optional BD FACSFlow™ supply system fluidics cart increases capacity and ease of use while maintaining a stable fluidics pressure. It includes an automated sheath and waste fluid control system that reduces daily maintenance by incorporating two 20-L containers (Cubitainers®).

Fluidic sensors maintain constant pressure, while a fluidics monitoring system warns when sheath fluid is low or empty, or when the waste container is full.



Hydrodynamic focusing of the sample core through the flow cell

Cuvette flow cell



Cuvette flow cell
A quartz cuvette flow cell is gel-coupled to the collection optics. This design helps ensure that lasers are precisely focused on the sample stream to generate the greatest signal and ensure that the maximum amount of emitted light is collected.

Maximum Signal, Minimum Crosstalk

An Innovative and Proven Platform for Multicolor Analysis

The patented collection optics are yet another design innovation. Arranged in octagon- and trigon-shaped optical pathways, their novel design efficiently maximizes signal detection and increases sensitivity and resolution. This allows researchers to identify cells, especially dim and rare cell populations, optimizing multicolor assays and panel design for superior results.

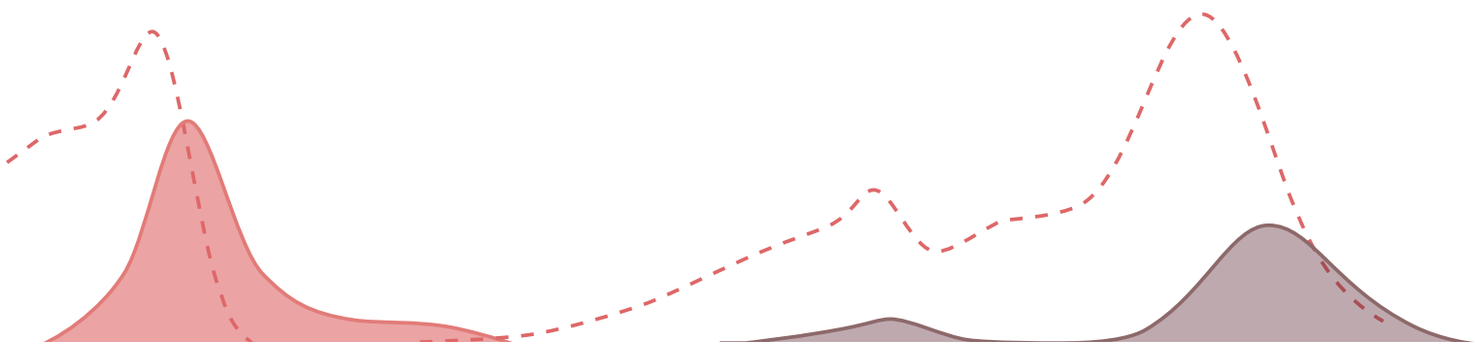
Optics System

The optics system consists of laser excitation optics that illuminate cells in the sample, and collection optics that direct light scatter and fluorescence signals through spectral filters to detectors. Innovative designs for both the excitation optics and collection optics in BD LSRFortessa systems reduce excitation losses and optimize collection efficiency for increased sensitivity and resolution.

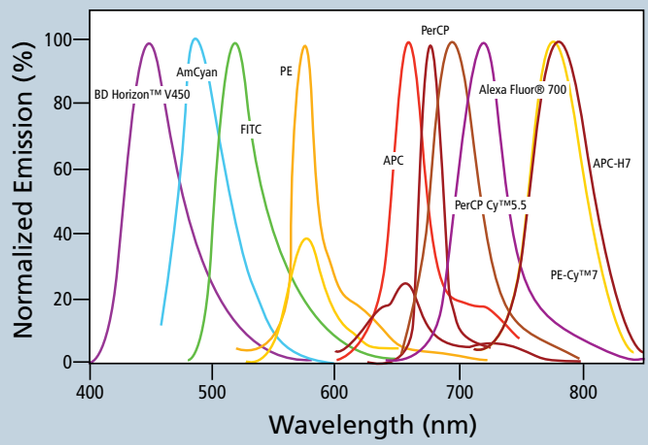
Excitation Optics

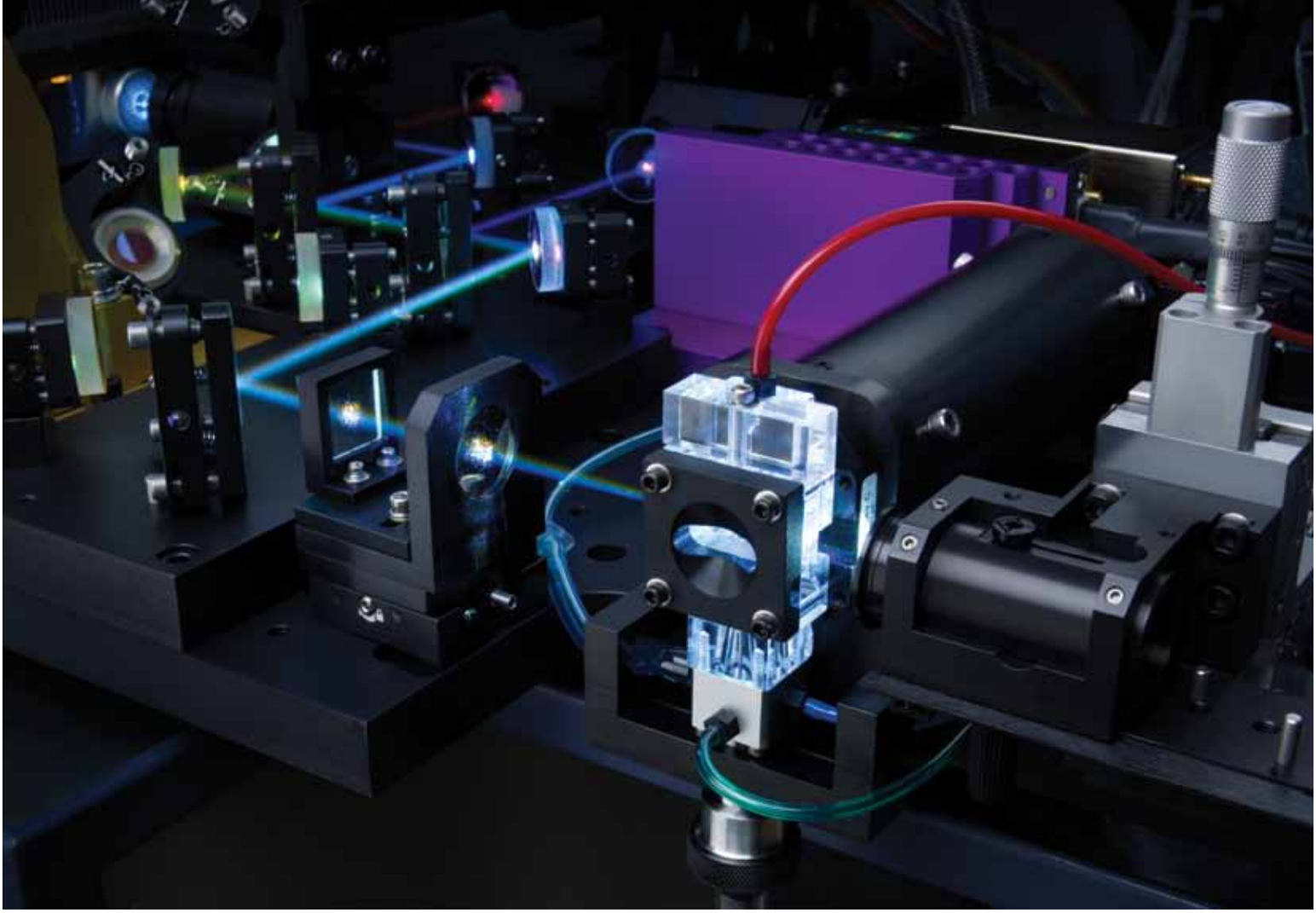
The excitation optics consist of multiple fixed wavelength lasers, beam shaping optics, and individual pinholes which result in spatially separated beam spots.

A final lens focuses the laser light into the gel-coupled cuvette flow cell. Since the optical pathway and the sample core stream are fixed, alignment is constant from day to day and from experiment to experiment.



Emission spectra of commonly used fluorochromes

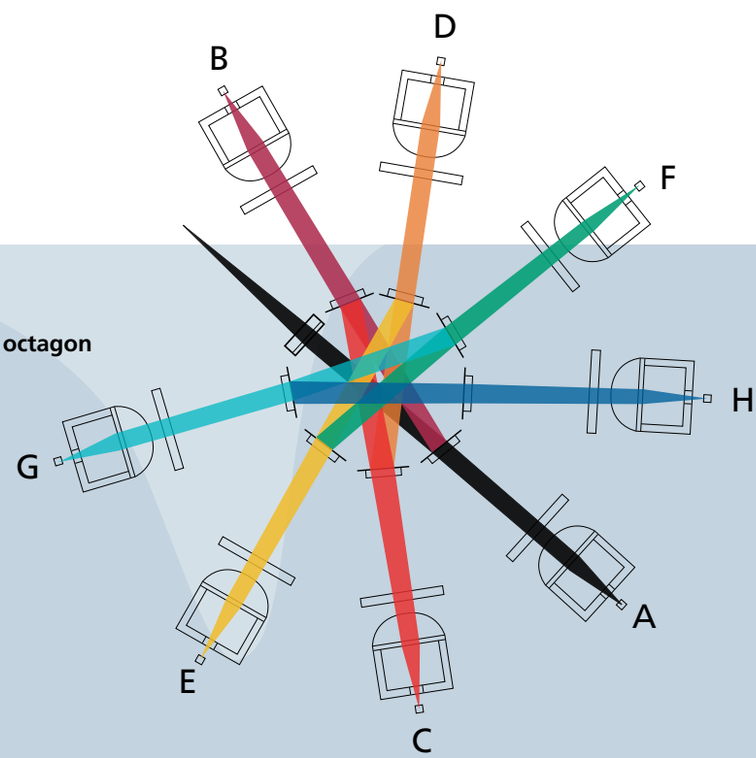




Collection Optics

Emitted light from the gel-coupled cuvette is delivered by fiber optics to the detector arrays. The collection optics are set up in patented octagon- and trigon-shaped optical pathways that maximize signal detection resulting from each laser illuminated beam spot. Bandpass filters in front of each PMT allow spectral selection of the collected wavelengths. Importantly, this arrangement allows filter and mirror changes within the optical array to be made easily and requires no additional alignment for maximum signal strength.

This design is based on the principle that light reflection is more efficient than light transmission. Emitted light travels to each PMT detector via reflection and is transmitted through only two pieces of glass to reach the detector. Therefore, more colors can be detected with minimum light loss.



Transmission pathways in an octagon

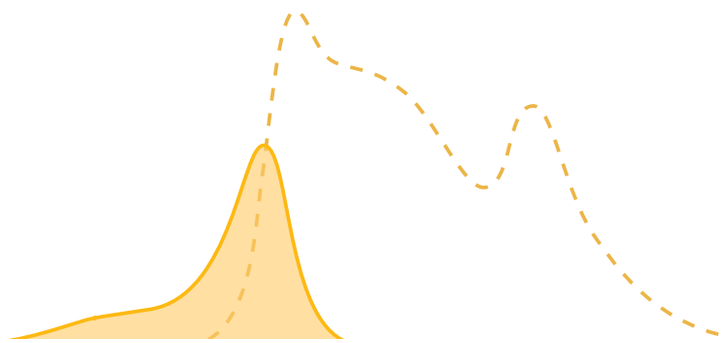
Greater Consistency

Automated Controls for Setup, Acquisition, and Analysis

BD FACSDiva™ software controls the efficient setup, acquisition, and analysis of flow cytometry data from the BD LSRFortessa workstation. The software is common across BD FACS™ instrument platforms, including the BD FACSCanto™ cell analyzer and BD FACSAria™ cell sorter systems. This affords researchers greater application flexibility, allowing them to easily move assays from one platform to another.

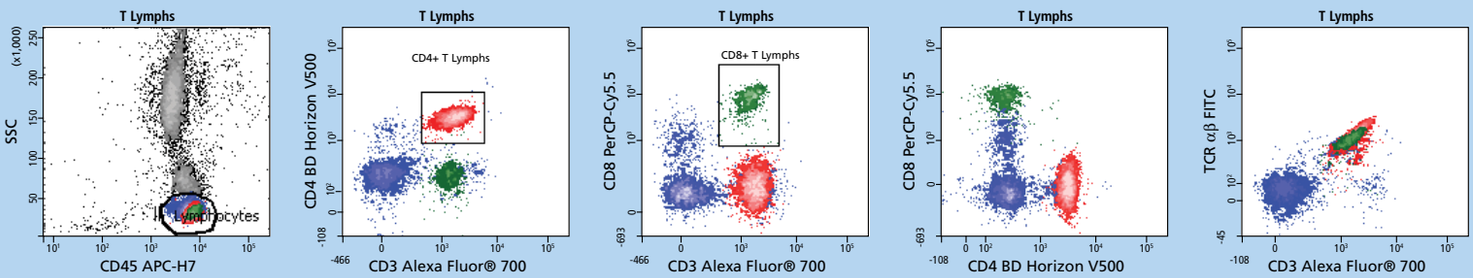
Cytometer Setup and Tracking

The Cytometer Setup and Tracking (CS&T) feature of BD FACSDiva software establishes baseline settings and adjusts for instrument variability. The software reduces operator error, and ensures consistency of results by setting the signal time delay across the multiple laser beams and optimizing PMT voltages. Application-specific settings can also be set, allowing for rapid setup and performance of routine experiments in a more consistent manner. Quality control (QC) tracking capabilities in the software measure instrument settings and report on performance. Levey-Jennings plots help users understand instrument performance and identify maintenance issues.



Analysis of human blood

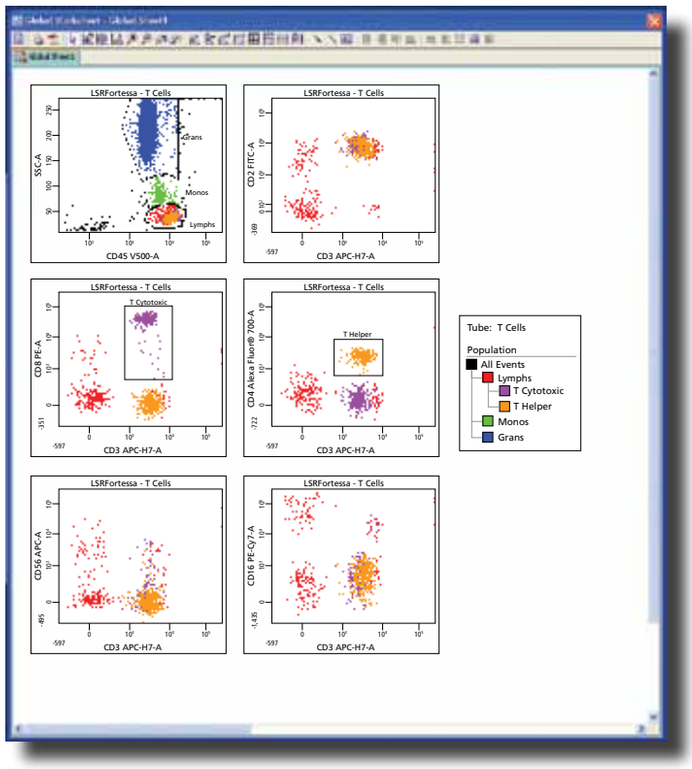
Data shows a 10-color panel run on a 3-laser BD LSRFortessa with a blue/red/violet configuration. The panel was used for defining T-cell subsets using lysed whole blood (BD Pharm Lyse™ lysing buffer) from a normal donor. Use of the BD Horizon™ V450 and BD Horizon V500 dyes allows brighter fluorochromes to be used for more dimly expressed antigens, while the use of 10 colors expands the amount of information gained from a single tube.



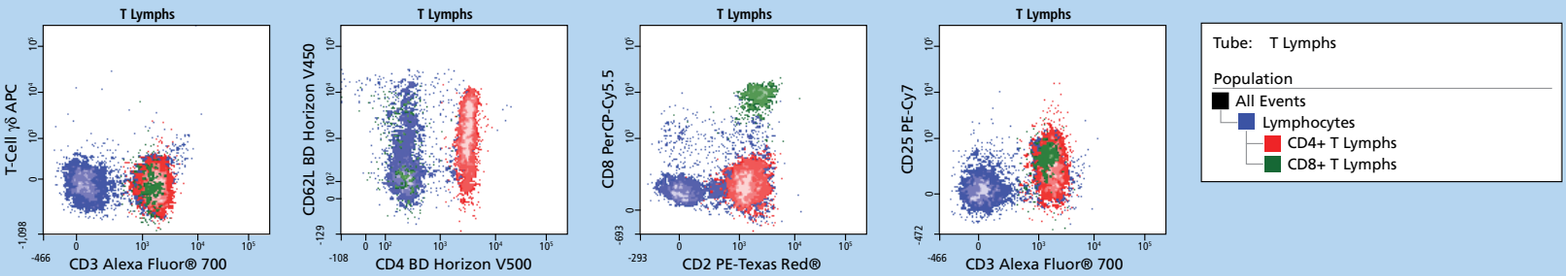
Acquisition and Analysis

BD FACSDiva software enables researchers to preview and record data from multiple samples with an automated acquisition process. Acquisition templates, user-defined experiment layouts, and simple compensation procedures are also managed by the software to further facilitate acquisition.

For analysis, the software includes powerful features such as hierarchical snap-to gating, a variety of plot formats, and batch analysis. Recorded data can be analyzed by creating plots, gates, population hierarchies, and statistics views on a BD FACSDiva global worksheet. Once the global worksheet is saved, it can be used to analyze multiple sample tubes from an experiment, thereby saving time. Numerous other productivity benefits include user-defined batch analysis and automated features such as gate resizing, pausing between data files, exporting statistics, and printing before proceeding to the next data file.



BD FACSDiva worksheet showing well defined T-cell population subsets acquired using the special order BD LSRFortessa analyzer configured with the following lasers: 60-mW 355-nm UV, 100-mW 450-nm violet, 100-mW 488-nm blue, and 40-mW 640-nm red.



Choices Now and in the Future

Special Order Products for Unique Needs

The BD special order program allows customers to configure BD flow cytometers and cell sorters to fit precise research and assay needs. Tailored to the special needs of research at the leading edge of biomedical discovery, the program offers a wide range of choices to help researchers create the ultimate customized instrument for their requirements.

Multiple Power Options

Innovative laser options include the choice of solid-state lasers across the full spectrum. Currently, 18 laser wavelength options are offered, ranging from ultraviolet to infrared wavelengths. Through the special order program, researchers can configure a BD LSRFortessa with up to 7 lasers and 42 positional choices for the detectors.

Forward Scatter PMT Option

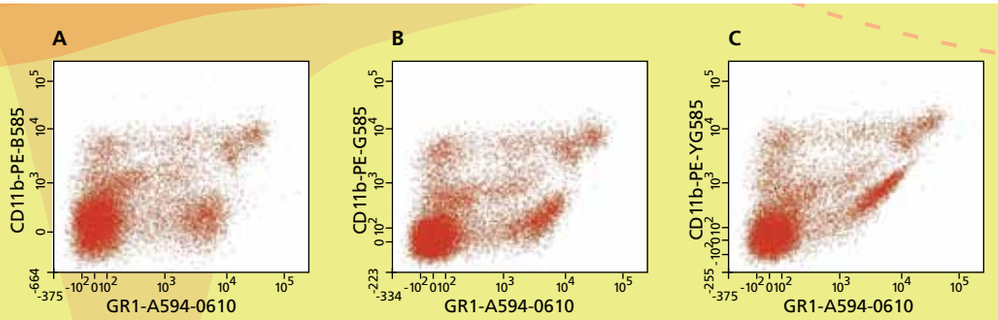
The special order program offers a Forward Scatter (FSC) PMT Option that complements small particle scatter detection. This option lowers the threshold on size measurements, which is particularly valuable for microbiology applications.

Evolving Capabilities

BD LSR customers have an expanded level of flexibility and choice because they can add lasers after purchasing a system, to meet new or evolving requirements. This flexibility makes BD LSR analyzers a solid, long term investment.

Optimization of Fluorochrome Excitation

These experiments demonstrate the versatility of the 10-laser special order BD LSR II cell analyzer. Providing the user choices for laser excitation wavelength, this system adapts to unique user defined fluorochromes, for optimization of multicolor experiments.



Effect of laser excitation on spectral overlap

Cells co-stained with Alexa Fluor® 594 (GR1) and PE (CD11b) were run using blue 488-nm (B), green 532-nm (G), and yellow-green 561-nm (YG) laser as alternate excitation sources for PE. Alexa Fluor® 594 was excited using an orange 592-nm laser. Panel A shows PE excitation with a 488-nm laser, panel B shows excitation with a 532-nm laser, while panel C shows excitation with a 561-nm laser. The 10-laser special order BD LSR II instrument allows the user to choose the excitation source as well as the excitation power. Researchers can maximize signal while minimizing the compensation required to deal with unwanted spectral overlap.

An Innovation Process

A vigorous pursuit of the latest and best laser technologies ensures an unparalleled range of configuration choices offered by the BD special order program for BD LSR analyzers. New technologies are regularly incorporated into the product line as soon as they become available.

The ever-expanding list of available lasers demonstrates BD's ongoing commitment to perpetual innovation. This is one of the many ways BD ensures that the BD LSR analyzer platform continues to support the evolving needs of leading researchers around the world.

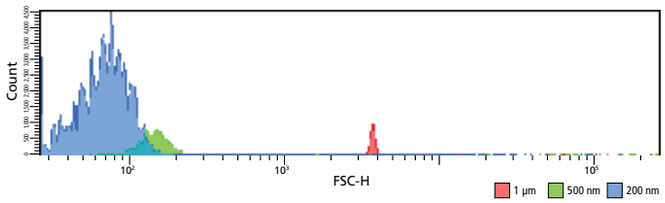


Figure 1: BD LSRFortessa Standard FSC

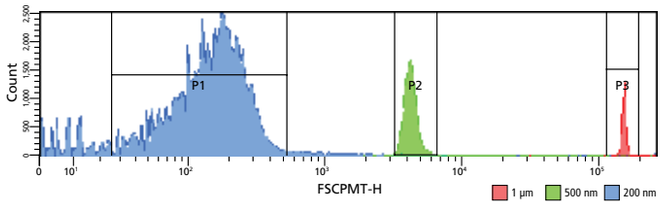
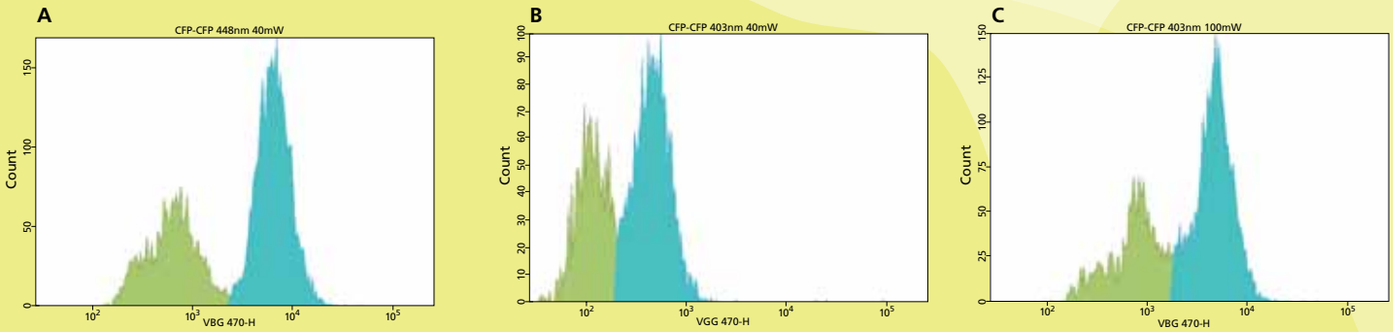


Figure 2: BD LSRFortessa Special Order FSC PMT Option

The BD LSRFortessa FSC (data shown in Figure 1) is optimized for a wide range of biological particles. For applications requiring smaller resolution, a FSC PMT Option is available through the special order program.



Excitation of ECFP using two different excitation wavelengths

Panel A shows data generated using a 448-nm excitation source at 40 mW, Panel B shows a 403-nm excitation source at 40 mW, and Panel C shows a 403-nm excitation source at 100 mW. While ECFP is optimally excited at 448 nm, it is apparent that an increase in laser power can largely compensate for a suboptimal excitation source. Green peak = ECFP-negative cells, aqua peak = ECFP-positive cells.

Data courtesy of A. Smith, R. Salomon, S. Allen, and B. Roediger of the Centenary Institute, University of Sydney, Sydney, Australia.

Services

BD Biosciences is fully committed to the success and satisfaction of its customers. Supporting flow cytometry applications for over 35 years, BD has training, technical applications support, and field service teams dedicated to helping customers achieve optimal instrument performance, ease of use, and streamlined workflow. With unmatched flow cytometry experience, this world-class service organization is available to help with your BD LSR product installation, future upgrades, and application support.

Training
Hands-on training is included with each BD LSRFortessa analyzer. Courses are held at BD training centers worldwide. BD flow cytometry training courses combine theory and practice to provide participants with the skills and experience they need to take full advantage of the capabilities of their BD LSRFortessa analyzer.

Technical Application Support
BD Biosciences technical applications support specialists are available to provide field- or phone-based assistance and advice. Expert in all aspects of flow cytometry, BD technical application specialists are well equipped to address customer needs in both instrument and applications support.

Field Service
When instrument installation or service is required, a BD Biosciences Technical Field Service Engineer can be dispatched to the customer site. BD Biosciences field service engineers are located across the world. On-site service and maintenance agreements are available to provide long-term support for BD LSR analyzers.

Special Order Program
Instruments can be customized to meet customer requirements via the special order program.

Regional Offices bdbiosciences.com/offices

Asia Pacific
Singapore
Tel 65.6861.0633
Fax 65.6860.1590
China
Tel 86.21.3210.4610
Fax 86.21.5292.5191
India
Tel 91.124.2383566
Fax 91.124.2383224/25/26

Australia/New Zealand
Australia
Toll Free: 1800 656 100
Tel 61.2.8875.7000
Fax 61.2.8875.7200
bd_anz@bd.com
New Zealand
Toll Free: 0800 572.468
Tel 64.9.574.2468
Fax 64.9.574.2469
bd_anz@bd.com

Europe
BD Biosciences
Tel 32.2.400.98.95
Fax 32.2.401.70.94
bdbiosciences.com/eu
help.biosciences@europe.bd.com

Canada
BD Biosciences
Toll Free 888.259.0187
Tel 905.542.8028
Fax 888.229.9918
bdbiosciences.com/ca
canada@bd.com

Japan
Nippon Becton Dickinson
Toll Free 0120.8555.90
Tel 81.24.593.5405
Fax 81.24.593.5761

United States
BD Biosciences
Toll Free 877.232.8995
Fax 800.325.9637
bdbiosciences.com
answers@bd.com

BD flow cytometers are Class I (1) laser products.

Cubitainer is a registered trademark of Hedwin Corporation.

Alexa Fluor® and Texas Red® are registered trademarks and Pacific Blue™ is a trademark of Molecular Probes, Inc.

Cy™ is a trademark of Amersham Biosciences Corp.

For Research Use Only. Not for use in diagnostic or therapeutic procedures.

Purchase does not include or carry any right to resell or transfer this product either as a stand-alone product or as a component of another product. Any use of this product other than the permitted use without the express written authorization of Becton, Dickinson and Company is strictly prohibited. Product availability and prices are subject to change without notice.

© 2011 Becton, Dickinson and Company. All rights reserved. No part of this publication may be reproduced, transmitted, transcribed, stored in retrieval systems, or translated into any language or computer language, in any form or by any means: electronic, mechanical, magnetic, optical, chemical, manual, or otherwise, without prior written permission from BD Biosciences.

BD, BD Logo and all other trademarks are property of Becton, Dickinson and Company. © 2011 BD

BD Biosciences
2350 Qume Drive
San Jose, CA 95131
bdbiosciences.com

23-11617-02