BD LSR II Flow Cytometer
Flexible • Expandable • Powerfull

BD Biosciences
Clontech
Discovery Labware
Immunocytometry Systems
Pharmingen
The BD™ LSR II flow cytometer is the most flexible yet powerful benchtop analyzer available. Free up your high-performance cell sorter by moving non-sorting applications that require a UV or Krypton laser to the BD LSR II.

Innovative technology in the BD LSR II optics and digital electronics have created a more sensitive flow cytometer that yields more information from each sample.
Flexibility Begins with Four Fixed-Aligned Lasers

The BD LSR II flow cytometer is fully configurable with one to four fixed-aligned, air-cooled lasers. By distributing a large number of fluorochromes over four lasers, you reduce spectral overlap and improve sensitivity. Fixed alignment provides consistent and stable day-to-day performance. The three new solid state lasers available on the BD LSR II flow cytometer—the Coherent® Sapphire™, Coherent VioFlame™ PLUS, and Lightwave Xcyte™—consume less power, produce less heat, and have a lower total cost over the life of the laser compared to gas-filled lasers.

The solid state Coherent Sapphire blue laser (20 mW @ 488 nm) produces no plasma glow, therefore signal-to-noise ratios are improved in 488 nm–excited fluorochromes.

The Coherent VioFlame PLUS violet laser (25 mW @ 405 nm) excites Cascade Blue®, Pacific Blue™, and Alexa Fluor® 405 dyes, once used only with expensive Krypton lasers. In addition, four-color fluorescent protein experiments are easily performed using BD Clontech Living Colors™ fluorochromes GFP, YFP, dsRed, and CFP with this laser.

The BD LSR II is the first commercially available flow cytometer to use a solid state UV laser. No special power or cooling requirements are needed, eliminating expensive room renovation costs. The Lightwave Xcyte laser (20 mW @ 355 nm) has outstanding performance on UV dyes such as DAPI, Hoechst, and Indo-1. UV DNA dyes, calcium flux, and stem cell side population experiments used to be run only on complex cell sorters. Now, with the benchtop BD LSR II flow cytometer, they are routine experiments.

With the addition of the HeNe laser (18 mW @ 633 nm), APC and APC-Cy7 dyes can be detected. The optical system also allows the simultaneous detection of Alexa Fluor 700 conjugates with APC and APC-Cy7.

Multiple laser, detector, and fluorochrome options make the BD LSR II flow cytometer system the most versatile benchtop analyzer on the market today.
Revolutionary new optics collects more light

The collection optics on the BD LSR II flow cytometer includes patented octagon and trigon optical arrays. The optical arrays increase the sensitivity and flexibility of the BD LSR II flow cytometer, yielding more information from each sample.

Light from four spatially separated laser beam spots is delivered by fiber optics to octagon and trigon detector arrays. The 488-nm octagon array is capable of collecting up to seven colors plus side scatter (SSC). The 633-nm, 405-nm and 355-nm trigon arrays can each detect up to three fluorescent colors. The concept behind the optical arrays is that reflectance of light is more efficient than transmittance. The optical arrays transmit the highest wavelength to the first PMT and reflect the lower wavelength to the next PMT through a series of longpass dichroic mirrors. More colors can be detected with minimum light loss.

The octagon and trigons have single-position insertion sockets for both inner reflectance mirrors and outer discrimination filters. Detecting more colors does not mean time-consuming adjustments; the BD LSR II flow cytometer allows filter and mirror changes to be made to the optical array without diminishing maximum signal.

The Power of Digital

Signal processing is powered by the digital acquisition system. Digital acquisition allows measurement of total fluorescence, thus providing more information per cell. The digital electronics continuously digitizes signals 10 million times per second into 16,384 discrete levels. The area measurement is the sum of all data points measured for each pulse, thereby making a direct measurement of total immunofluorescence possible over a 4.2-decade range. Logarithmic amplifiers and analog peak-and-hold circuits are no longer required. By eliminating the log amps and dead time, and using a digital virtual zero baseline restore algorithm, more accurate fluorescent measurements are possible. Linearity, compensation, and fluorescent quantitation are improved.

Rare event analysis is more efficient when events are acquired at a faster rate. Faster digital processing allows the BD LSR II flow cytometer to acquire and process events at a rate of greater than 20,000 events per second with minimal electronic aborts.
High-Performance BD FACSDiva Software

BD FACSDiva™ software drives experiment design, instrument control, and data analysis on the BD LSR II system. The powerful PC-based BD FACSDiva software improves workflow with features such as reusable acquisition templates, an auto-spillover matrix for automated compensation, offline compensation, and Biexponential data display, which allows better visualization of digital data. Data, instrument settings, experiment plots, compensation matrices, and statistics are stored in a database for quick reanalysis or for re-use in future experiments.

<table>
<thead>
<tr>
<th>Instrument</th>
<th>Laser</th>
<th>Excitation Laser Line (nm)</th>
<th>Fluorescence Channel</th>
<th>Fluorochromes from BD Biosciences</th>
</tr>
</thead>
<tbody>
<tr>
<td>BD LSR II Flow Cytometer (typical setup)</td>
<td>Blue</td>
<td>488</td>
<td>Green</td>
<td>FITC, PE, Alexa Fluor® 488, ECFP</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Yellow</td>
<td>PE</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Red</td>
<td>PerCP</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Infra Red</td>
<td>PE-Cy7</td>
<td></td>
</tr>
<tr>
<td>Red</td>
<td>633</td>
<td>Red</td>
<td>APC*</td>
<td>Alexa Fluor® 647</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Infra Red</td>
<td>APC-Cy7</td>
<td></td>
</tr>
<tr>
<td>UV</td>
<td>355</td>
<td>Violet</td>
<td>Alexa Fluor® 350</td>
<td></td>
</tr>
<tr>
<td></td>
<td>405</td>
<td>Blue</td>
<td>Pacific Blue®</td>
<td></td>
</tr>
</tbody>
</table>

* APC and PE-Cy5 may be used together on instruments with cross-beam compensation.
Options to increase productivity

Different options can tailor the BD LSR II flow cytometer to your laboratory’s specific needs. The BD LSR II flow cytometer is able to sample from a 96- or 384-microtiter tray with the BD™ High Throughput Sampler (HTS). Based on sampling speed or strict sample carryover tolerances, the HTS offers adjustable parameters through a software user interface. This then allows the BD LSR II flow cytometer to be used as a high-content, high-speed screening instrument. Additional PMTs and electronics can also be added, for detection of up to 14 colors.

How to Get the Most out of the BD LSR II Flow Cytometer

BD Customer Education provides comprehensive training on BD instruments and software. Small classes offer personalized instruction on the proper use and maintenance of the BD LSR II flow cytometer, as well as how to best use BD FACSDiva software on the BD LSR II flow cytometer for maximum performance.

BD Worldwide, Worldclass Service

The BD LSR II flow cytometer is backed by BD worldwide phone and field service team. BD FACService™ support guarantees response time, and provides preventative maintenance programs and highly trained service and application specialists to answer any of your questions.

For more information on the BD LSR II Call your BD Biosciences Sales Representative.

Figure B

Using an eight-color panel for antigen-specific phenotyping, informational content is maximized providing possible evidence of novel patterns of T-cell differentiation among CMV-responsive donors. Figure B1 shows a sequential gating strategy for phenotyping of IFN_ positive cells after CMV simulations. Figure B2 are the results of total CD8 T cells from a CMV+ donor. Biexponential scaling is used in all fluorescent parameters to avoid data distortion produced by purely logarithmic displays. Data courtesy of Laurel Nomura, BD Biosciences San Jose, CA

Figure C

The plots show a calcium flux experiment with CD4 and CD8 surface marker staining of PBMCs. The cells were activated by the addition of purified CD3 and measured over time. Indo-1 AM was excited by the 355-nm Xcyte UV laser.