



Helping all people
live healthy lives

BD FACS Aria Flow Cytometer

The first high-speed sorter with a fixed-alignment
cuvette flow cell from BD Biosciences

From Pioneers to Revolutionaries in Flow Cytometry Instrumentation

BD Biosciences continues to provide the most advanced instrumentation in flow cytometry. Since 1974 when we developed the first commercially available fluorescence activated cell sorter (FACS™) in collaboration with Stanford University, BD Biosciences has been the leader in flow cytometry technology and innovation. BD Biosciences continues to be the industry leader with a revolutionary new high-speed cell sorter, the BD FACSAria™ instrument. This novel instrument platform has been created from the ground up with over 25 years of experience in flow cytometry instrumentation. Our technological advances in flow cytometry help customers advance their research and discovery.

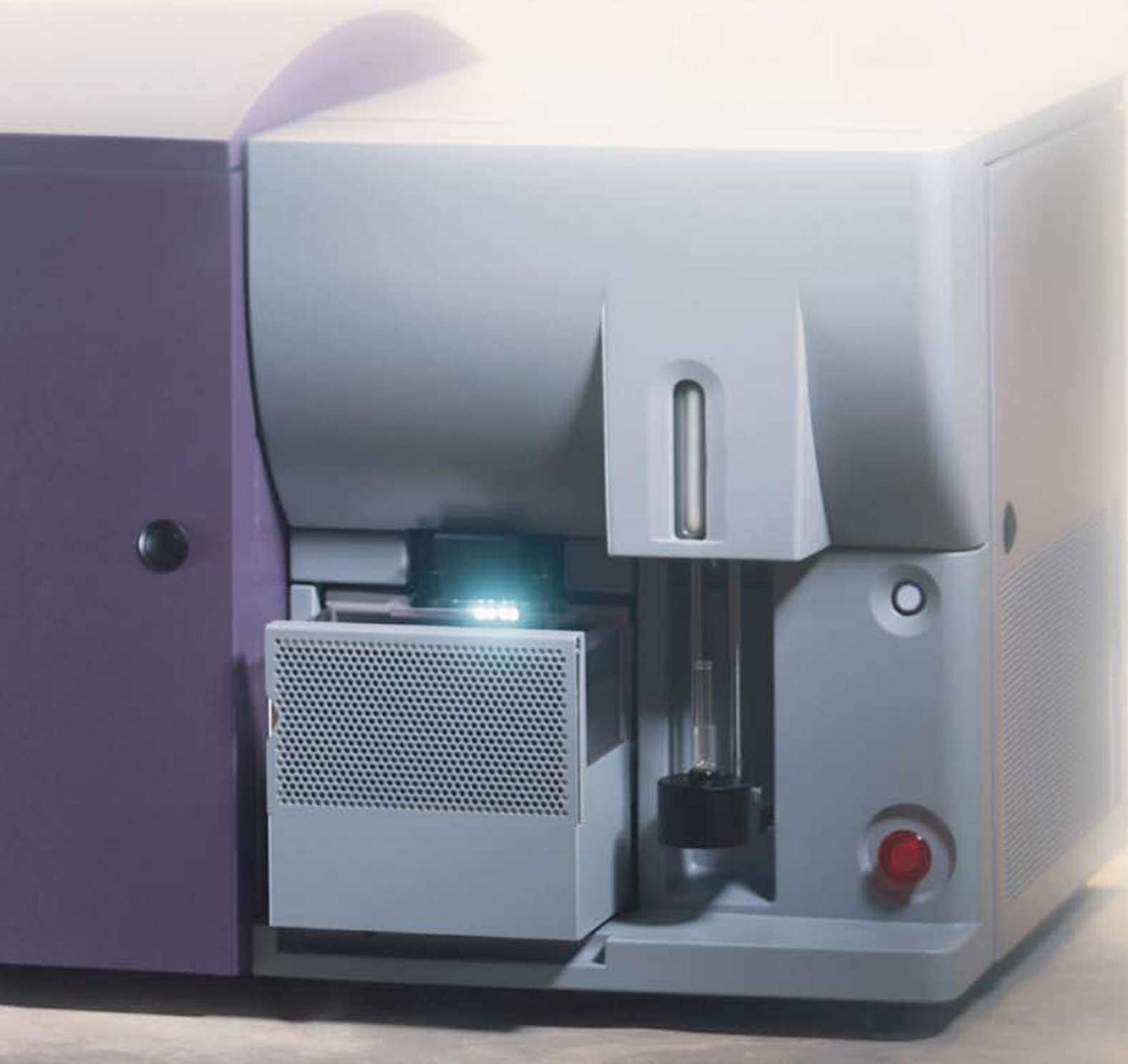
The BD FACSAria cell sorter is named for the musical term *Aria*, meaning a striking solo performance. The BD FACSAria cell sorter embodies this name because it is able to perform independently without the burden of special room requirements or years of technical expertise. It is easy to operate and provides superior performance in high-speed cell sorting and multicolor analysis.

BD Biosciences is committed to helping each laboratory excel scientifically by providing the most advanced technology in flow cytometry. The reliability and performance of our instrumentation, reagents, service, and support have been key to customer satisfaction. BD Biosciences offers the complete solution in flow cytometry.



Next Generation Flow Cytometry High-Speed Sorter

The BD FACSAria cell sorter sets a new standard for high-performance flow cytometry. Based on an entirely new design in instrumentation, the BD FACSAria instrument is the first benchtop sorter that incorporates a fixed-alignment cuvette flow cell. The numerous technological advances embodied in the BD FACSAria cell sorter reduce the cost of owning a high-speed sorter and accelerate research by providing unparalleled, easy-to-use, high-performance sorting and analysis.



The Heart of the BD FACSaria Cell Sorter

The cuvette flow cell is the heart of the BD FACSaria cell sorter. New technologies incorporated in the instrument design create a truly fixed-alignment optical path. The fixed optical alignment frees the operator from tedious instrument optimization, allowing the focus to center on the science rather than the instrument.

The cuvette flow cell achieves high sensitivity in signal detection with low-powered lasers, allowing air-cooled and solid state lasers to be utilized in this system. This, in turn, eliminates the need for special power and cooling requirements associated with high-powered lasers used in conventional stream-in-air sorters.

Fiber optics direct and focus the laser light in a precise and constant manner on the sample core stream. Fiber optics steer the three lasers, 488 nm, 633 nm, and 405 nm, onto the alignment prisms, and then the lasers are focused on the cuvette flow cell (see Figure 1). Because the placement of the sample core stream within the cuvette flow cell and the laser alignment are fixed, daily instrument optimization of the cytometer is no longer necessary.

Two major advantages of the cuvette flow cell design are improved light excitation and collection optics. Excitation by the BD FACSaria cell sorter differs from a stream-in-air sorter. The sample passes through the laser beams at a lower velocity, allowing longer exposure to laser energy. The stream is accelerated only as it enters the nozzle tip. The drop drive then breaks the stream into droplets to be sorted.

Another advantage of the BD FACSaria cell sorter is how it collects light signals. Stream-in-air sorters can overcome excitation losses by increasing laser power. However, at some point, most fluorophores saturate. Only a better collection system can increase sensitivity. A gel-coupling cuvette flow cell collects at least four times the amount of light over stream-in-air sorters. The BD FACSaria instrument is the only high-speed sorter with a gel-coupled cuvette, thus improving sensitivity in signal detection (<125 Molecules of Equivalent Soluble Fluorescein [MESF]).* See Figure 10 on page 9 for a demonstration of the high sensitivity achieved with the cuvette flow cell.

*Using SPHERO™ Rainbow Calibration Particles RCP-30-5A

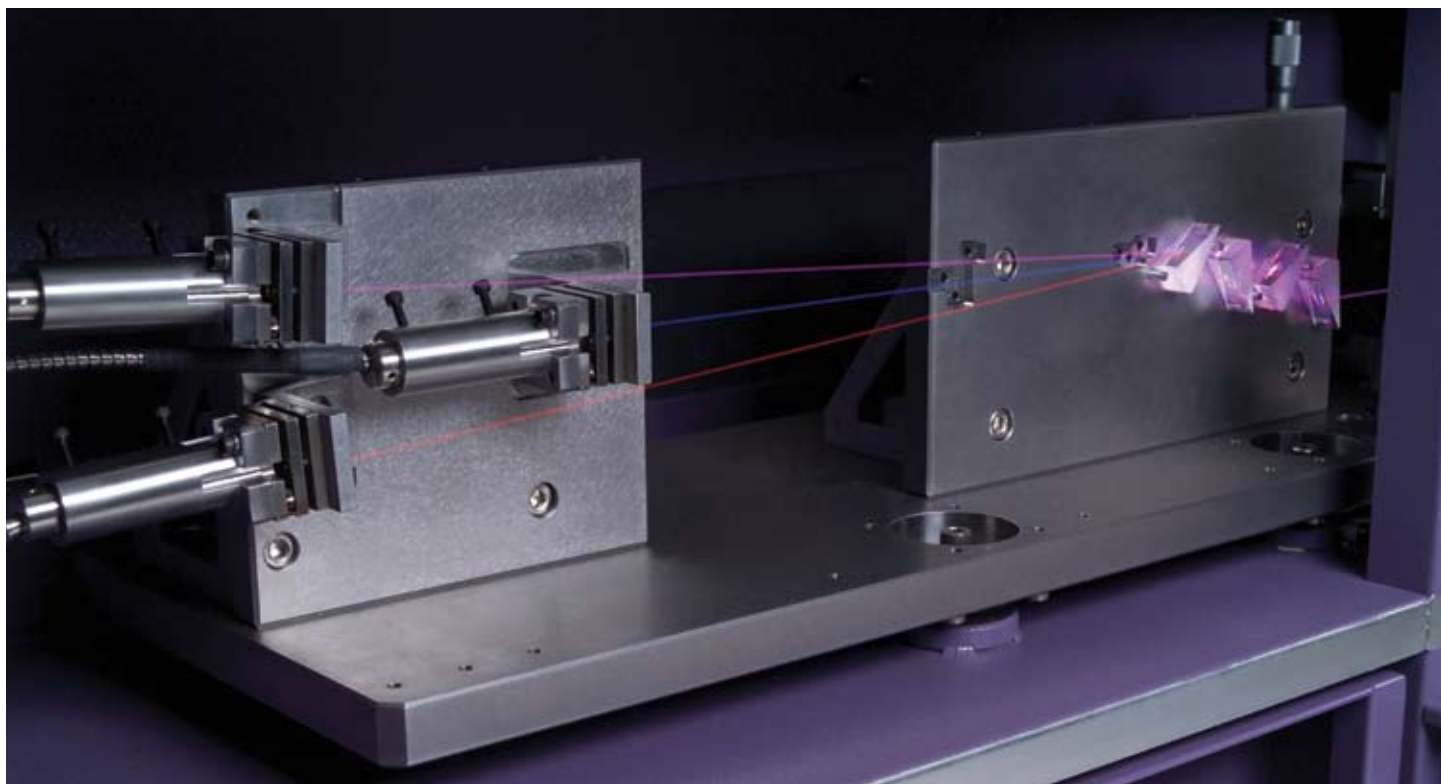


Figure 1
Excitation optics focus the 488-nm, 633-nm, and 405-nm laser beams

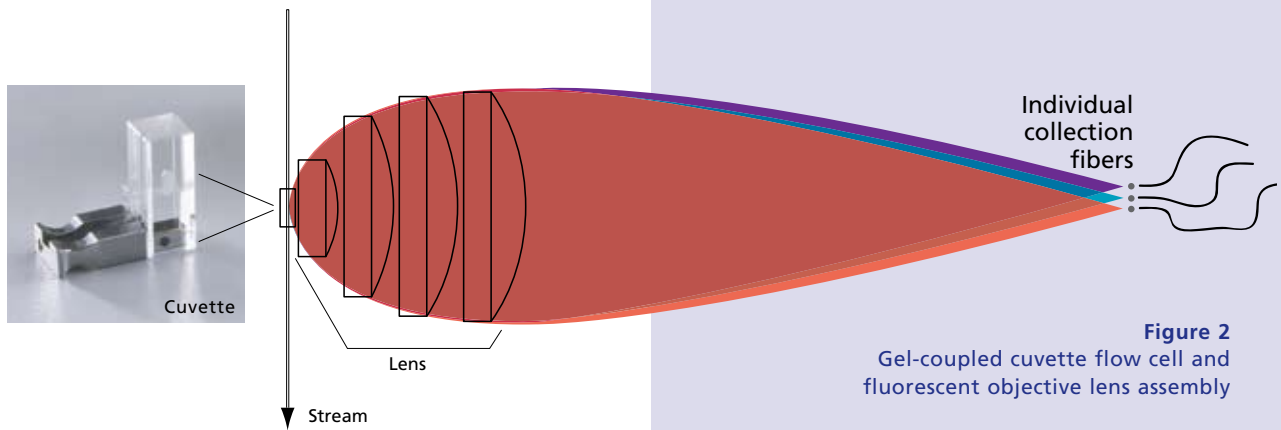


Figure 2
Gel-coupled cuvette flow cell and
fluorescent objective lens assembly

Revolutionary Optics System

The cuvette flow cell is gel-coupled to the fluorescent objective lens to transmit the greatest amount of light to the collection optics (see Figure 2). The fluorescent objective lens collects and focuses the fluorescent light emitted at each of the three laser focal points onto individual collection fibers. The collection fibers then transfer the emitted light onto the collection optics. The collection optics are engineered to achieve the greatest amount of signal detection from each laser. This is accomplished by transmitting the highest wavelengths to the first PMT and reflecting the lower wavelengths to the next PMT through a series of longpass dichroic mirrors. Bandpass filters in front of each PMT allow fine-tuning of the collected signal. Since reflection is more efficient than transmittance, this design greatly increases the multicolor detection capabilities of the instrument (see Figure 3).

The collection optics consist of octagon- and trigon-shaped collection devices. The octagon-shaped collection device for the 488-nm laser detects up to seven fluorescent colors plus side scatter. The trigon-shaped collection devices for the 633-nm and 405-nm lasers each collect up to three fluorescent colors. With the addition of FSC, a total of 15 independent signals can be acquired at one time. This entirely new optical system increases the quality and quantity of the information acquired from each sample. Representative data is shown in Figures 11 and 12 on page 9.

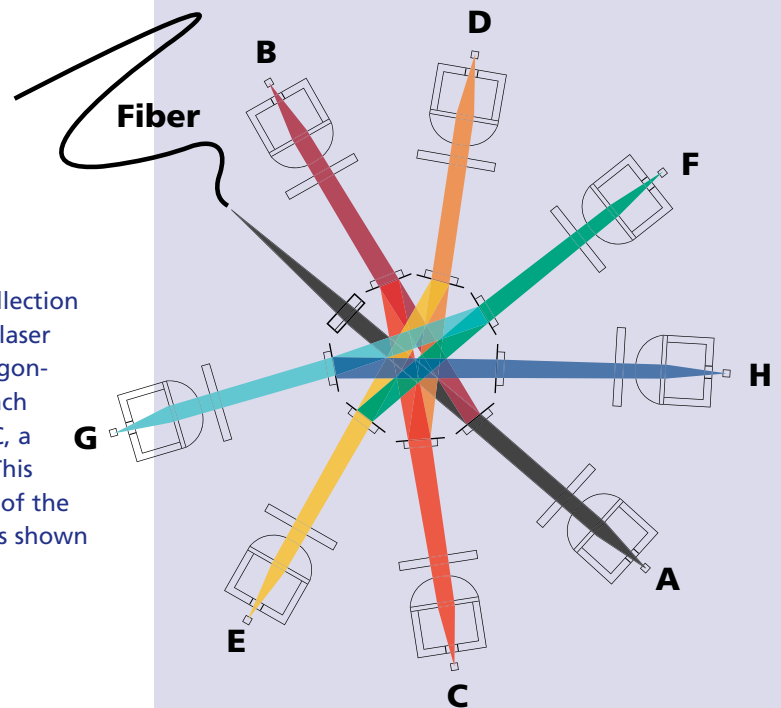


Figure 3
Octagon-shaped collection optics



Figure 4
Fluidics cart



Figure 5
Plenum reservoir

Technological Advances in the Fluidics System

The fluidics system is another key technological advancement embodied in the BD FACS Aria system. The fluidics system includes an entirely new means of delivering both the sample and the sheath fluid to the instrument. A new fluidics cart supplies the sheath and cleaning fluids from non-pressurized containers and collects the waste from the instrument.

The self-contained fluidics cart supplies air pressure and vacuum eliminating the need for an external source. This feature allows the instrument to operate in any lab or room without the special and costly facility requirements of a traditional high-speed flow cytometer. The air pump provides pressure from 2–75 psi (14–517 kPa) to accommodate a variety of sorting applications. The fluidics cart holds four 10-L containers, two sheath and two waste, and three 5-L auxiliary cleaning fluid containers (see Figure 4). The cart also accommodates BD FACSFlow™ sheath fluid containers.

The fluidics system transfers sheath fluid from the fluidics cart into a plenum reservoir inside the BD FACS Aria system (see Figure 5). The plenum reservoir holds the sheath fluid before it is pumped into the cuvette flow cell and removes any pressure changes as the plenum reservoir is refilled. Thus, pressure is no longer dependent on the level of fluid in the sheath container, and the reservoir removes small air bubbles from the sheath supply. The entire fluidics system is designed to further enhance this easy-to-use instrument.

The instrument has a specialized sample injection chamber that accommodates different tube sizes to provide flexibility in experimental design (see Figure 6). Once a tube is placed in the tube holder of the sample injection chamber, the entire chamber becomes pressurized. This allows even cracked tubes to be sampled and aids sample containment. The tube holder agitates the sample, keeping cells constantly suspended throughout a long sort.

Because the sample injection chamber is self-contained, specialized cleaning modes are available. After each tube is removed, sheath fluid flushes the sample tubing inside and out to eliminate potential sample carryover. Using a special cleaning mode, the entire sample injection chamber can be filled with ethanol.

Figure 6
Sample injection chamber



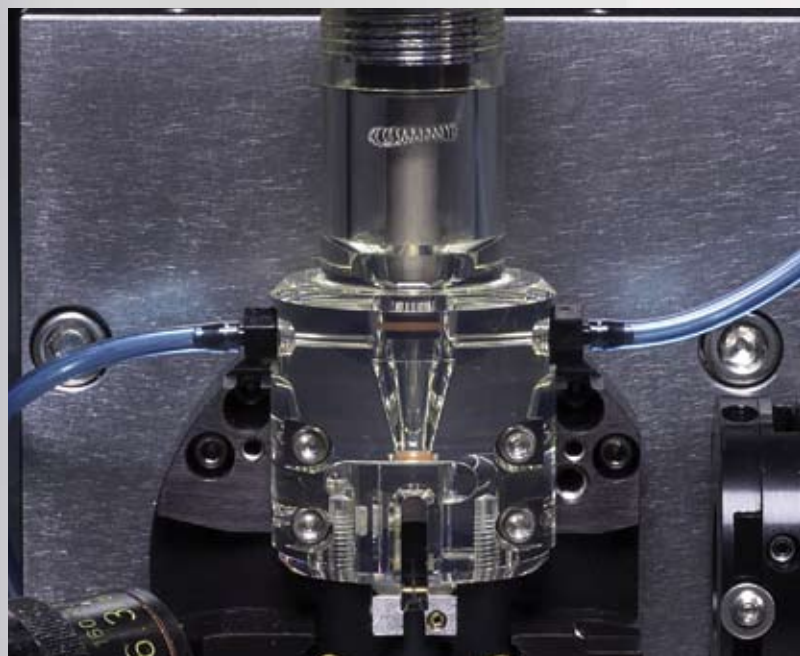
New Era for High-Speed Cell Sorting

With the BD FACS Aria cell sorter, high-speed sorting has never been easier to set up and perform. Advances in instrument design give the BD FACS Aria system improved sort performance without sacrificing results. Due to the unique design of the cuvette flow cell, drop drive disturbances that create noise on stream-in-air systems are not seen on the BD FACS Aria cell sorter. Unlike stream-in-air systems, there is no perceptible performance degradation of any parameter while using the drop drive. Thus, data quality is not compromised during sorting.

The nozzle tip is keyed into a fixed position at the end of the cuvette ensuring reproducible stream alignment into the waste aspirator (see Figure 7). Fixed nozzle alignment ensures the stream will return to the same spot after a nozzle tip is removed and reattached.

The 70- and 100-micron nozzle tip sizes are designed to accommodate most cell types. The nozzle sizes enable sorting at a variety of pressures and speeds. Two- and four-way sorting are standard features available for a variety of bulk collection devices to accommodate different tube sizes (for example, microtubes, 12 × 75 mm, and 15 mL). The optional automated cell deposition unit (ACDU) sorts into multiwell plates and onto microscope slides (see Figure 14 on page 11). The flexibility of the BD FACS Aria cell sorter provides the total solution for flow cytometry sorting.

Figure 7
Cuvette flow cell and nozzle tip



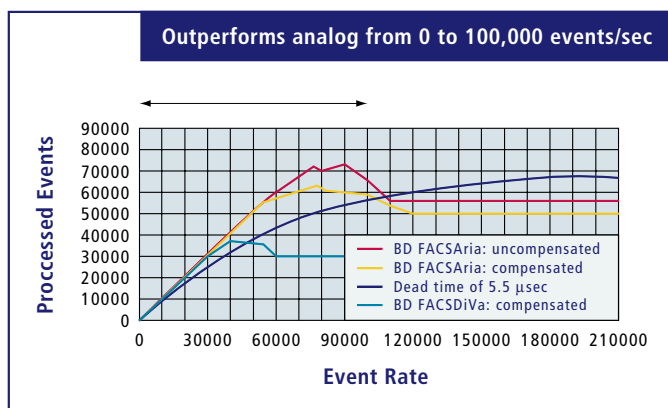


Figure 8
BD FACSAria system acquisition rates

Advanced Digital Electronics Achieve Unparalleled Performance

Advanced digital electronics improve sort performance and provide significant advantages in instrument ease-of-use. The electronics eliminate dead time. This improves the number of signals that can be processed and sorted, thus increasing sort yield by reducing the number of electronic aborts. The electronics simplify compensation of all fluorescent signals with no limitation of inter- and intra-beam compensation.

Faster signal processing enables acquisition rates of up to 70,000 events per second for 15 parameters. The BD FACSAria system can achieve faster acquisition rates than an analog system. See Figure 8 for eight-parameter data generated for instrument comparison.



Figure 9
BD Aerosol Management Option

Aerosol Management

While sample is acquired and sorted, the sample injection and sort collection chambers are enclosed to help with aerosol management. The sort collection chamber is completely sealed and operates under negative pressure from the waste aspirator. To reduce the chance of biohazard exposure, the instrument rinses the sample tubing between samples.

The BD™ Aerosol Management Option (AMO) (see Figure 9) is an optional device that provides additional aerosol management and removal within the sort chamber area. The AMO is equipped with a 0.01-μm pore size ultra-low penetrating air (ULPA) filter to trap aerosolized particles. The AMS evacuates the volume of the ACDU chamber and sort area 16 times per minute in normal evacuation mode and 69 times per minute in rapid evacuation mode.

Representative Data

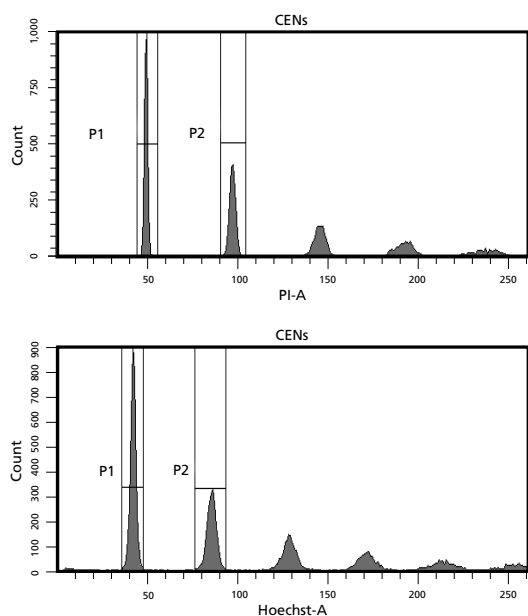


Figure 10
DNA stained with PI and Hoechst

The high sensitivity of the BD FACSaria cell sorter is shown in Figure 10. In DNA stained with PI and Hoechst acquired at 70 psi and 90 kHz, CEN stained with PI resulted in a CV of 1.5% and with Hoechst a CV of 3.5%.

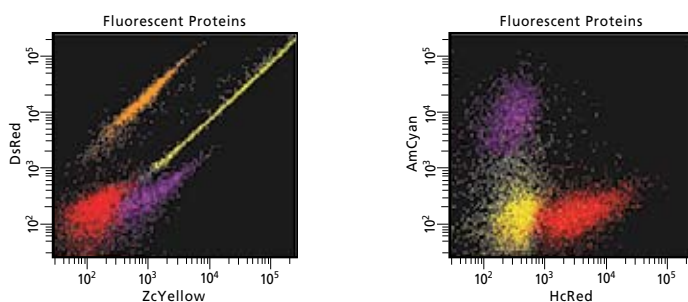


Figure 11
Fluorescent proteins

Its revolutionary optics allow the BD FACSaria instrument expanded possibilities in multicolor analysis. In Figure 11, four unique fluorescent proteins from BD are excited by the 488-nm, 633-nm, and 405-nm lasers: AmCyan, DsRed, ZcYellow, and HcRed.

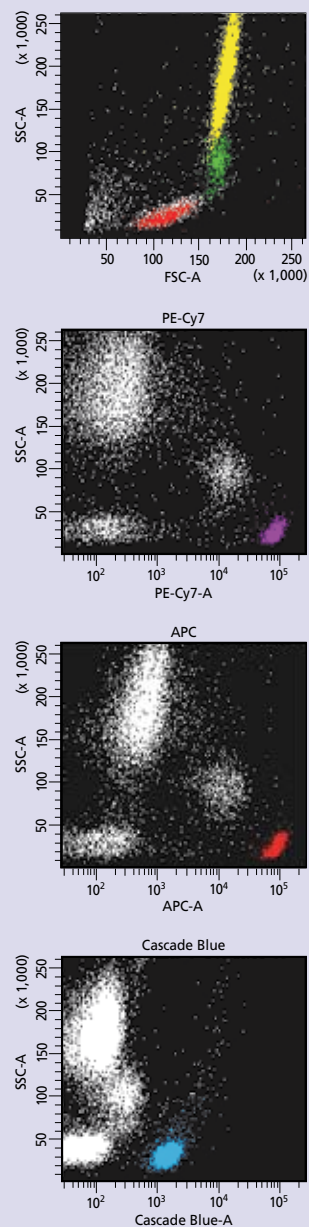


Figure 12
Whole blood stained with CD4 single-color fluorophores

The BD FACSaria cell sorter achieves high separation between negative and positive events as shown in Figure 12. In normal whole blood stained with CD4 single-color fluorophores and acquired at 70 psi and 90 kHz, signal is detected from each laser: PE-Cy™7 from the 488-nm laser, APC from the 633-nm laser, and Cascade Blue® from the 405-nm laser.

Increased Sorting Advances Through Software

BD FACSAria™ software provides unparalleled instrument control for flexible sorting and data analysis. All instrument and sorting controls are embedded in the software to simplify instrument setup and operation. New features programmed into the software provide the complete solution for high-performance cell sorting.

Fixed-nozzle alignment and a software breakoff monitoring system ensure that the drop delay value does not change unless a nozzle is replaced. Integrated BD FACS™ AccuDrop technology allows the user to determine an accurate drop delay value. See Figure 13 for a software display of stream breakoff and side streams.

Comprehensive sort monitoring and clog detection are standard integrated software features that make the BD FACSAria cell sorter easy to use. The breakoff monitoring system—referred to as the *sweet spot* in recognition of Dr. Richard Sweet who holds the patent on this technology—automatically adjusts the amplitude to maintain the same breakoff value throughout a sort. The clog detection feature signals the instrument to stop the sort and protect the collection tubes if the instrument cannot maintain the stream breakoff.

Increased drop resolution provides accurate investigation of each sorted drop and results in better sort purity and yield. Each drop is investigated in 32 increments. By examining the sorted drops in smaller increments, each sort decision is accurate and efficient. Increased resolution increases sort accuracy and performance by sorting only the drops of interest.

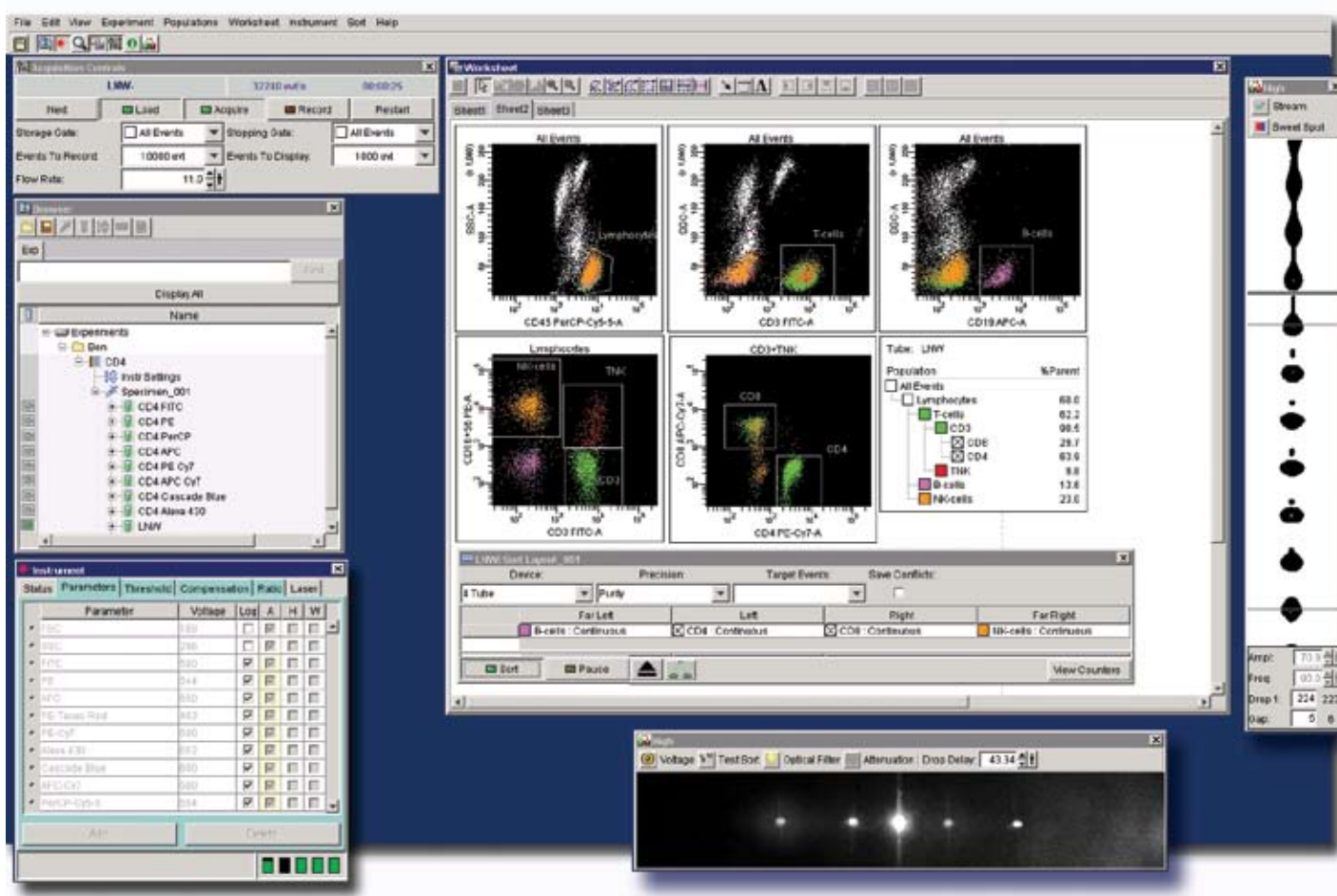


Figure 13
Software display of stream breakoff and side streams

BD FACSAria Workstation and Software

The BD FACSAria system operates on a PC-based Windows® platform. The system includes a variety of removable storage devices and comes equipped with a high-resolution LCD flat-panel monitor. Ethernet is built in for networking capabilities. A three-port FireWire® PCI adapter is also provided. A wireless keyboard and mouse provide freedom of movement and flexibility while performing sample optimization and sort setup.

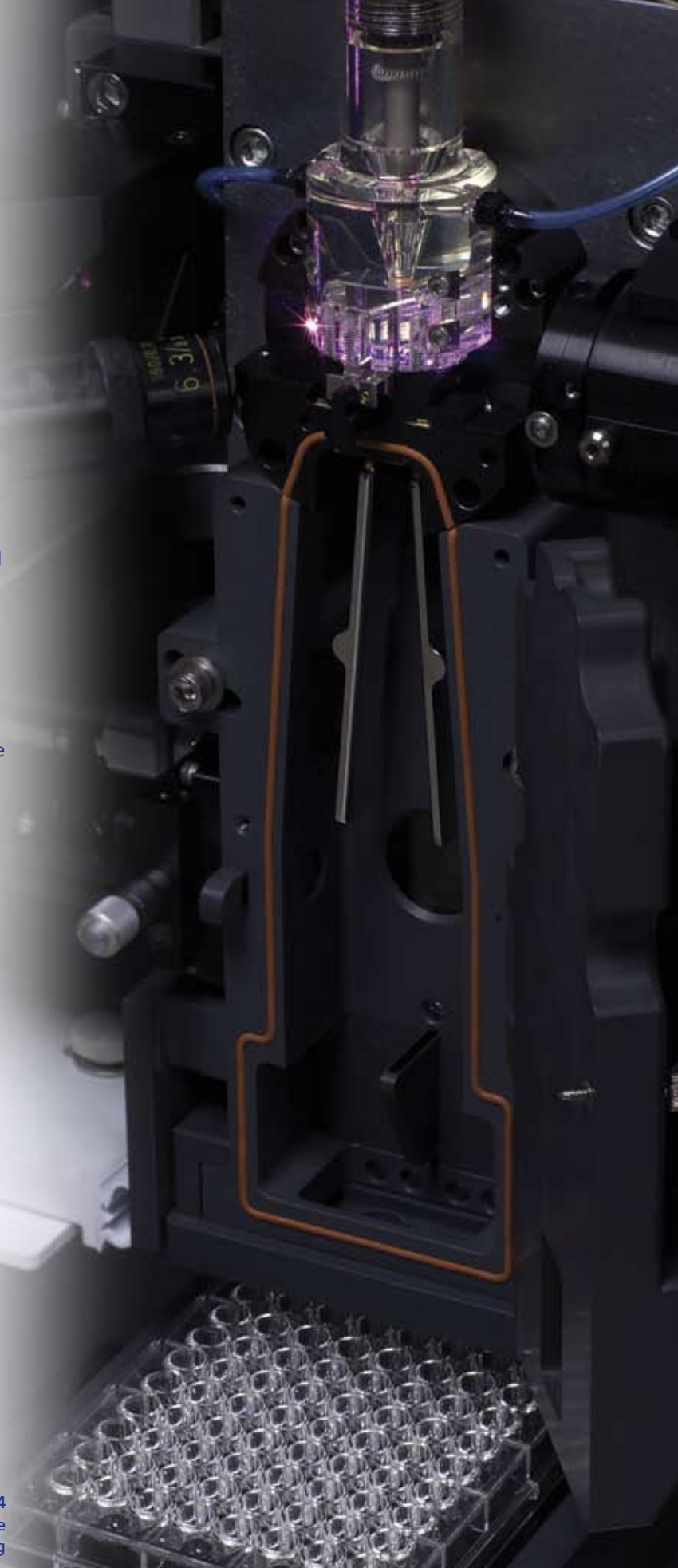
Unique Design

The BD FACSAria instrument was engineered to be space efficient and compact with reduced overall size compared to conventional sorters. The instrument can be installed and operated in virtually any room without special facility requirements. The instrument can be set up on a typical laboratory benchtop or table and requires only a standard electrical outlet.

Large water-cooled lasers are no longer necessary to achieve high fluorescent sensitivity and sorting performance. The fluidics system is supported entirely by the self-contained fluidics cart. The BD FACSAria instrument is revolutionary in all aspects of its design and operation.



Figure 14
ACDU option allows plate
and slide sorting



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