BD FACSAria™ III Cell Sorter

The BD FACSAria™ III cell sorter is built on the solid foundation of patented technologies, multicolor performance and renowned ease-of-use that has led to the unparalleled success of the BD FACSAria™ cell sorter portfolio. Since its introduction in 2003, each successive generation of the BD FACSAria has opened the complex world of cell sorting to a broader audience of researchers and wider range of applications. Now, the BD FACSAria III system is even more powerful, dependable and easier to use.

The BD FACSAria III delivers proven multicolor performance. Its fluidics and optical systems include unique design innovations precisely integrated to maximize signal detection. These innovations include the laser excitation optics, the patented flow cell with gel-coupled cuvette and the highly efficient, patented octagon and trigon modules. Together, these systems allow the BD FACSAria III to achieve unrivaled sensitivity and resolution.

The BD FACSAria III is powerful and expandable. The system can mount up to five lasers, so you can choose the configuration that meets application, budget and site requirements. An innovative new X-mount optical plate makes this possible. It accommodates easy expansion to six lasers and four spatially separated beam spots. Wavelength choices include 488-nm, 633-nm, 405-nm, 561-nm and 355-nm lasers. Mount up to 20 detectors and measure a maximum of 18 colors simultaneously.

BD

Technical Specifications

Optics

Excitation optics

Optical platform

Fixed optical alignment of all Class IIIb lasers upon the cuvette flow cell. The 488-nm and 633-nm lasers come standard. All other laser choices are optional. All are solid state except the 633 nm, which is gas.

Beam height: 9 ±3 µm
Beam width: 65 ±7 µm

Power out of the laser head

355 nm: >15 mW, elliptical shape
405 nm: >50 mW, elliptical shape
488 nm: >20 mW, elliptical shape
561 nm: >50 mW, elliptical shape
633 nm: >18 mW, elliptical shape

Emission optics

Steering optics

488, 633, 561 and 405-nm laser: Fiber optics steer the fixed alignment laser beams onto the expansion prisms to focus them on the cuvette flow cell.

UV (355-nm) laser: Air-launched and focused on the cuvette flow cell at the same location as the 405-nm lasers.

Optical coupling

The quartz cuvette flow cell is gel-coupled by refractive index-matching optical gel to the fluorescent objective lens for optimal collection efficiency.

Numerical aperture: 1.2

Forward scatter detector and filters

Photodiode with 488/10 bandpass filter for the 488-nm laser.

Side scatter detector

Photomultiplier with a 488/10 bandpass filter for the 488-nm laser.

Fluorescence detectors and filters

Four fixed-fiber apertures. The 488-nm, 633-nm and 561-nm lasers have their own aperture and fiber. The 405-nm and 355-nm lasers share an aperture and fiber.

An octagon technology detector array enables user-defined detection configurations.

Filters and mirrors are user-changeable. Additional detectors up to a total of 18 wavelengths can be added to the arrays.

Wavelengths detected from 488-nm laser without 561-nm laser installed

530/30-nm FITC
585/42-nm PE
616/23-nm PE-Texas Red®
695/40-nm PerCP-Cy™5.5 or PI or 675/20-nm PerCP
760/60-nm PE-Cy™7

Wavelengths detected from 488-nm laser with 561-nm laser installed

530/30-nm FITC
695/40-nm PerCP-Cy5.5 or PI or 675/20-nm PerCP

Wavelengths detected from 633-nm laser

660/20-nm APC
780/60-nm APC-Cy7 or APC-H7

Wavelengths detected from 405-nm laser

450/50-nm BD Horizon™ V450, Pacific Blue™, DAPI
510/50-nm BD Horizon V500, AmCyan

Wavelengths detected from 561-nm laser

582/15-nm PE
610/20-nm PE-Texas Red®, Living Colors®, mCherry, propidium iodide
670/14-nm PE-Cy™5 or 710/50-nm PE-Cy5.5
780/60-nm PE-Cy7

Wavelengths detected from 355-nm laser

450/20-nm Hoechst Blue, DAPI
670 LP Hoechst Red, PI

Fluidics

General operation

The fluidics cart provides sheath and cleaning fluids to the instrument and receives waste. No air or vacuum required. Room air can be used if desired. Sheath pressure is adjustable from 5 to 75 psi.

Fluidic reservoirs

Autoclavable 10-L sheath and waste containers and 5-L cleaning reservoirs provided.

Sample flow rates

Adjustable dynamic range of sample flow rates.

Fluidic cleaning modes included (software)

Automated startup and shutdown
Clean flow cell
Prepare for aseptic sort

Nozzles

70-, 85-, 100- and 130-µm, removable and can be sonicated.

A registered key-fit position at the bottom of the cuvette provides fixed stream alignment.

Sample collection cooling/heating

Refrigerator/heater option is available to provide cooling or heating for sort collection into tube holders, multiwell plates and slides.

Operating temperature range:
-20°C to 120°C

Automated cell deposition unit (ACDU)

ACDU for slide and plate sorting: 6-, 24-, 48-, 96- and 384-well plates.

Performance

Fluorescence sensitivity
Measurements performed at 70 psi and 90 kHz using SPHERO™ Rainbow Calibration Particles (RCP-30-5A)

FITC: 85 molecules of equivalent soluble fluorochrome (MESF-FITC)

PE: 29 molecules of equivalent soluble fluorochrome (MESF-PE)

Fluorescence resolution
Coefficient of variation (CV)
PI: Area, <3.0%, full G_0/G_1 peak for propidium iodide (PI)-stained chicken erythrocyte nuclei (CEN)

Hoechst: Area, <3.5%, full G_0/G_1 peak for Hoechst-stained CEN

Fluorescence linearity
Doublet/singlet ratio: CEN stained with PI: 1.95–2.05 (488-nm laser) or Hoechst: 1.95–2.05 (405-nm laser)

Forward and side scatter sensitivity
Sensitivity enables separation of fixed platelets from noise, identification of bacteria and detection of 0.5-µm beads.

Forward and side scatter resolution
Scatter performance is optimized for resolving lymphocytes, monocytes and granulocytes.

Sample acquisition rate
Maximum acquisition rate (events per second) with 12 compensation pairs and 8 parameters: 70,000.

Sort performance

Drop drive frequency
Range 1–100,000 Hz

Purity and yield
At 70 psi and 90 kHz with an average threshold rate of 25,000 events per second, a four-way sort achieved a purity of >98% and a yield >80% of Poisson's expected yield. Higher threshold rates up to 70,000 events per second can be achieved without affecting purity; however, yield will decrease based on Poisson statistics.

Viability
Sorts were performed at a variety of sheath pressures using cell lines and human peripheral blood mononuclear cells (PBMCs). All sorts resulted in cells that proved viable and proliferated for several days post-sort.

Sort collection devices
Two-way sorting: 12 x 75 mm, and 15 mL
Four-way sorting: 1.5 mL microtube and 12 x 75 mm

Sort collection cooling
Water recirculator for refrigeration/heating (optional)

BD FACS™ Accudrop
Red diode laser provided for fully automated drop-delay determination
Automated drop breakoff monitoring
Automated clog detection and sort tube protection system using Sweet Spot technology

Signal processing

Converter
10-MHz Analog-to-Digital converter. Pulse sampling is precisely matched to particle flow rate in the cuvette. Particles travel slower compared to conventional stream-in-air sorters. This increases the light collected, resulting in better sensitivity. High-speed sorting is achieved by accelerating the stream through the nozzle, achieving drop rates comparable to stream-in-air sorters. The flow cell design and electronics are matched to maximize signal while maintaining maximum sort speed, purity and yield.

Workstation resolution
262,144 channels

Data acquisition channels
20 parameters, 18 fluorescent and 2 scatter

Fluorescent compensation
No limit to inter- and intra-beam compensation

Pulse processing
Height, area and width measurements available for any parameter. Ratio measurements are also available.

Time
Time can be correlated to any parameter for kinetic experiments or other applications.

Channel threshold
Available for any parameter from any lasers with the ability to use multiple thresholds from different lasers simultaneously.

Loading

Sample input sizes
Microtubes, 12 x 75 mm and 15 mL
Polystyrene or polypropylene tubes can be used.

Sample input agitation
Adjustable through the software to keep sample constantly suspended

Temperature control
Sample input, software-adjustable: 4, 20, 37 and 42°C

Sample output for sort collection: water recirculation unit (optional)

Data management

Workstation
PC workstation with at least Pentium® 4 processor, 3.0 GHz or faster

Memory
>2 GB RAM

Data storage
80-GB and 250-GB hard drives
16x DVD +/- RW, dual layer
Floppy drive

Networking
10/100/1000 Ethernet
FireWire® serial bus

Monitor
Two 19-inch LCDs, 2560 x 1024 resolution (standard)

One 20-inch LCD, 1600 x 1200 resolution (optional)

One 23-inch LCD, 1920 x 1200 resolution (optional)

Printer
Networkable color-laser printer

Data file structure
Flow Cytometry Standard (FCS) 3.0 or 2.0

Software
BD FACSDiva™ software v6.1.3 or later

Installation requirements

Instrument Dimensions
BD FACSAria III sensor (cell sorter):
28 H x 28 D x 48 W in. (71 x 71 x 122 cm)
400 lb (181.4 kg)

BD FACSAria III fluidics cart (wet cart):
26 H x 26 D x 32 W in. (66 x 66 x 81 cm)
180 lb (81.7 kg)

See Site Preparation Guide for space and clearance requirement.

Temperature operating range
Between 17.5°C (63.5°F) and 27.5°C (81.5°F)

Heat dissipation
5100 BTU per hour

Power
Operation at 100/115/230 VAC and 50 or 60 Hz

Maximum power 1,500 watts

Water supply
None required

Air supply
None required

Table (optional)
BD FACSAria III instrument and computer table: 33.5 H x 53 D x 105 W in. (85 x 135 x 267 cm) 100 lb (45.4 kg)

Options

Aerosol Management Option (AMO)
The BD FACSAria III features an enclosed pathway from the sample injection chamber to the sort collection tubes. For an added level of aerosol management, the BD Aerosol Management Option (AMO) evacuates the sort collection chamber and traps aerosolized particles during sorting. It is equipped with a 0.01-µm size ultra-low penetrating air (ULPA) filter to trap aerosolized particles. Evacuates the volume of the ACDU chamber area 16 times per minute in normal evacuation mode and 69 times per minute in rapid evacuation mode.

When operated under normal and stressed conditions (mimicking a clog), <3 Glo Germ™ particles were identified outside the ACDU sort collection chamber. Glo Germ particles, developed by Glo Germ in Moab, Utah, have been shown to provide good visualization of aerosol deposition in normal and mock failure modes by Oberyszyn and Robertson (Cytometry. 43:217-222, 2001).

Available lasers for upgrade
355 nm
405 nm
561 nm

BD optical filters for BD Horizon Brilliant™ polymer conjugates available for the BD FACSAria™ Family.

Regulatory status

CE marked for electrical safety (Europe)
UL Standard for Safety Electrical Equipment for electrical safety (USA)
CSA for electrical safety (Canada)
Class I (1) laser product per CDRH regulations and EN/IEC 60825

Class 1 Laser Product.
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BD Life Sciences, San Jose, CA, 95131, USA

bdbiosciences.com