BD Influx Cell Sorter

Technical Specifications

The BD Influx™ cell sorter is a flexible flow cytometry platform that adapts to a researcher's application requirements. A modular architecture and a combination of detection capabilities, hands-on controls, and high performance allow researchers to configure the BD Influx system to various site and application needs.

The BD Influx system can handle a throughput rate of up to 200,000 events per second.

Optics

Excitation Optics

Optical Platform
Lasers are mounted on a standard optical breadboard. Laser beams are aligned independently by adjustable mirrors. Each laser beam has its own final focus lens mounted on a dedicated translational stage for fine adjustments. For systems with more than five lasers, the laser paths have one or more collinear beams through the steering optics and focusing lens, but intercept the stream at up to seven spatially separated points.

Lasers
A selection of laser wavelength and power is available. New laser combinations may require custom engineering of the optical platform.

Power Out of the Laser Head
355 nm: >100 mW, >250 mW
405 nm: >100 mW
445 nm: >100 mW
457 nm: >300 mW
488 nm: >200 mW
515 nm: >100 mW
532 nm: >150 mW
561 nm: >75 mW, >150 mW
594 nm: >100 mW
640 nm: >120 mW
785 nm: >40 mW

Beam-shaping optics provide elliptical beam spots for selected laser paths (3:1 ratio with a typical beam height of 15–20 µm).

Emission Optics

Emission light is collected through a 20X, 0.6 NA microscope objective (90°). Light is focused on three, five, or seven spatially separated mirror pinholes depending on the number of lasers. Modular detector blocks allow for a user-defined detection configuration (see the *BD Influx Cell Sorter Filter Guide*).

Simultaneous video observation of the stream, the pinholes, and the laser intercepts allows for fast and intuitive alignment.

Regular forward scatter is detected using 75- and 50-mm lenses, an aperture, and a photomultiplier tube. Resolution for the standard forward scatter detector is >0.5 µm (measured using beads). Collection angle is 2–17°.

Side scatter is collected through the 90° collection lens and measured using a photomultiplier tube. Side scatter resolution is >0.2 µm (measured using beads and 0.1-µm filtered sheath fluid).

Fluidics

General Operation
- Laboratory air pressure and/or vacuum can be used for operation (regulated at 90 psi, 6.2 bar).
- An optional air pressure supply and/or vacuum pump is available.
- Sheath pressure is adjustable from 1–90 psi (0.07–6.2 bar).

Fluidics Reservoirs
Autoclavable 7-L sheath and waste containers, equipped with pressure and vacuum readout, are provided.

Fluidics Control
- Sheath, sample, and boost pressure can be individually adjusted.
- A sample flow fine adjustment is provided for precise regulation of sample flow.
- Purge, pulse, rinse, and run buttons are provided for quick stream startup and bubble removal.

Replaceable Fluidics Path
- The fluidics path, including the nozzle assembly, can be exchanged. There are no inline valves. Only pinch valves are used.
- The sample line can also be exchanged.

Bubble Detector
A bubble detector in the sample line detects air bubbles from the sample tube and stops sample flow when the sample tube is empty, preventing air bubbles from reaching the nozzle assembly.

Sample Input
12 x 75-mm tubes, polypropylene

Temperature Control
Sample tubes can be cooled or heated by an optional circulating water bath.

Performance

Fluorescence Sensitivity
Measured using SPHERO™ Rainbow Calibration Particles (RCP-30-5A) according to the manufacturer's specifications:

Sheath pressure:	33 psi
Drop dive:	On, ~68 kHz
Excitation:	488 nm, 200 mW
Emission:	530/40 nm for FITC 580/30 nm for PE

FITC: 125 molecules of equivalent soluble fluorochrome (MESF-FITC)
PE: 125 molecules of equivalent soluble fluorochrome (MESF-PE)

Fluorescence Resolution
Measured using propidium iodide (PI)–stained chicken erythrocyte nuclei (CEN):

Sheath pressure:	33 psi
Drop drive:	On, ~68 kHz
Excitation:	488 nm, 200 mW
Emission:	610/20 nm for PI

Coefficient of variation (CV) of PI: <3%, full G_0/G_1 peak

Fluorescence Linearity
Measured using propidium iodide PI-stained CEN:

Sheath pressure:	33 psi
Drop drive:	On, ~68 kHz
Excitation:	488 nm, 200 mW
Emission:	610/20 nm for PI

Doublet/singlet ratio: 1.95–2.05

Sort Performance

Drop Drive Frequency
Adjustable 9–180 kHz

Purity and Yield
At 60 psi and 100 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 for all four populations. Higher threshold rates of up to 70,000 events per second can be achieved without affecting purity; however, yield will decrease based on Poisson statistics.

Viability
As shown in published literature, sorts performed using murine[1-4] and human[5] cells and/or cell lines[6] demonstrated good recovery and viability in several experimental systems. Optimal sort conditions need to be established for different cell types.

Nozzles
Supplied: 70, 86, 100, and 140 µm

Optional: 200 µm

Sort Collection Devices
All collection devices are designed to fit on the Computerized Cell Deposition Unit (CCDU). The CCDU is standard on all instruments.

- Two-way sorting: Microtubes; 12 x 75-mm, 15-mL, and 50-mL tubes
- Three-way sorting: One 50-mL tube and two 12 x 75-mm tubes
- Four-way sorting: Microtubes, 12 x 75-mm tubes
- Plates and slides: 6, 24, 48, 96, and 384-well plates; slides; and user-defined collection devices

Sample Collection Cooling
Water recirculator for refrigeration or heating of collection tubes (optional).

Sort Monitoring
- Live video feed of waste collection and side streams.
- Live video feed of breakoff point.
- Drop-delay determination is achieved with BD FACS™ Accudrop technology. The drop-delay value can be adjusted while viewing BD Accudrop beads which are illuminated by a red diode laser in the center and side sort streams.

Signal Processing

Data Acquisition Channels
- Up to 5-laser systems: 16 channels, usually 14 colors plus forward and side scatter
- Systems with 6 lasers or more: 24 channels, usually 22 colors plus forward and side scatter

Signal Processing
- 16-bit analog-to-digital conversion, 65,536 channels
- Parallel data stream with channel ID and integrity check
- Less than 1 correlation error per 10^8 events

Acquisition Rate
Dead time is 0 µs. The maximum throughput rate is 200,000 events per second, independent of the number of parameters.

Fluorescence Compensation
- Up to 5-laser systems: 16 x 16 digital compensation matrix.
- Systems with 6 lasers or more: 24 x 24 digital compensation matrix.
- Compensated parameters are being added to the bus as separate parameters.

Pulse Processing
- All signals are height (peak) by default.
- Pulse processor electronics add area and width measurements for a maximum of 8 parameters to the bus.
- Width measurement on the trigger parameter is standard.

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

Channel Threshold
- Any parameter can be used as the threshold from the primary laser.
- Lasers and detectors can easily be switched to change laser sequence.

Workstation

Workstation
PC workstation with Intel® Quad Core (3.06 GHz or faster), Windows® 7 Professional 64-bit operating system

Memory
6 GB RAM

Data Storage
- 2 x 500 GB hard drives, RAID 1 (mirrored) configured
- 16x CD/DVD +/- RW
- 2 USB 2.0

Networking
10/100/1000 Gigabit Ethernet

Monitor
20-inch LCD, 1280x1024 resolution

Data File Structure
Flow cytometry standard (FCS) 3.0

Software
BD FACS™ Sortware v1.0 or later

Installation Requirements

Dimensions (H x D x W)
Cytometer: 72 x 48 x 62 in.
(183 x 122 x 158 cm)

Cytometer with HEPA-filtered enclosure:* 86 x 50 x 77 in.
(218 x 127 x 196 cm)

Operational footprint (D x W):
84 x 84 in. (214 x 214 cm)

*A minimum of 102.4 in. (260 cm) floor-to-ceiling height is required during installation of the HEPA-filtered enclosure.

Weight
Cytometer: 229 lb (104 kg)

Cytometer with HEPA-filtered enclosure: 441 lb (200 kg)

Temperature Operating Range
59–77°F (15–25°C)

Heat Dissipation
6,200 BTU/h maximum, dependent on laser choice

Electrical Requirements
North America: 120 ± 10% VAC, 50/60 Hz, 15 A

Outside North America: 230 ± 10% VAC, 50/60 Hz, 10 A

Japan: 100 VAC, 50/60 Hz, 15 A

BD requires two dedicated circuits for the cytometer and computer system with a dedicated AC source not shared with any other equipment. Multiple peripherals may require an additional line. An appropriate transformer will be delivered depending on region-specific settings. Optional air and vacuum supply require an additional line.

Power
Power requirements are between 1,080 watts (3 lasers, no HEPA-filtered enclosure) and 1,800 (5 lasers with HEPA-filtered enclosure). Standby status is not applicable.

Humidity
55% ±10% RH

Noise
<80 dBA from all running equipment

Air Supply
90 psi (6.2 bar) regulated. The source of compressed air must deliver clean (less than 5 ppm), dry-filtered (oil-free) air and stable pressures.

Vacuum Supply
A vacuum supply is delivered with the system. A laboratory vacuum supply between 5 and 15 in. Hg at 1 CFM can also be used.

Options

Small particle

Small particle with polarization

HEPA-filtered enclosure

Aerosol evacuation

Sample temperature control

Polarization (without small particle)

Air compressor

Regulatory Status

CE marked for electrical safety (Europe)

UL Standard for electrical safety (USA)

CSA for electrical safety (Canada)

Class I (1) laser product per CDRH regulations and EN/IEC 60825-1

References

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4. Seamons A, Perchellet A, Goverman J. Endogenous myelin basic protein is presented in the periphery by both dendritic cells and resting B cells with different functional consequences. *J Immunol.* 2006;177:2097-2106.
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6. Panchision DM, Chen HL, Pistollato F, Papini D, Ni HT, Hawley TS. Optimized flow cytometric analysis of central nervous system tissue reveals novel functional relationships among cells expressing CD133, CD15, and CD24. *Stem Cells.* 2007;25:1560-1570.

Class I (1) laser product.

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

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