



**SH800S** Cell Sorter

Sony Biotechnology Inc.



### SH800S Cell Sorter

#### Sorting Made Simple™

The benchtop SH800S cell sorter permits sorting of a wide range of cell sizes for many applications using the 70- $\mu$ m, 100- $\mu$ m, and 130- $\mu$ m microfluidic sorting chips. This novel, chip-based design is fully integrated with comprehensive fluidic controls and advanced automation for setup, acquisition, sorting, and analysis to make sorting less subjective, more precise, and easier to perform.

System software is intuitive and supports sorting into tubes and 96- and 384-well plates. The software generates FCS 3.0 and FCS 3.1 files that can be exported to third party analysis tools.

The SH800S is flexible and can support a variety of applications for individual and core labs. For applications in which cross-contamination is a concern, optional e-beam sterilized consumables can be used to replace the sample line and sorting chip.

An optional class II type A2 biosafety cabinet provides protection for personnel and products. The biosafety cabinet was custom designed for the SH800S and tested by The Baker Company to meet many international standards.

The optical design offers up to four collinear excitation lasers (488 nm, 405 nm, 561 nm and 638 nm) and six fluorescence detectors. The six free-form PMTs enable detection of fluorescence signals from any laser, based on filter selection.



- Provides the highest level of automation available in any cell sorter, allowing researcher setup, alignment, calibration, and monitoring with a push of a button.
- A novel microfluidic sorting chip is available in three sizes including 70- $\mu$ m, 100- $\mu$ m, and 130- $\mu$ m to permit sorting of a wide range of cell sizes, which simplifies sorting.
- An optional custom biosafety cabinet that meets industry standards for personnel and product protection.



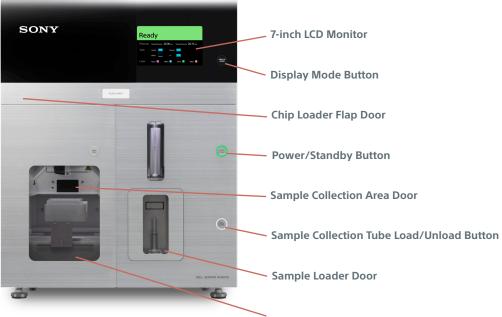
The SH800S features a compact footprint (55 cm W x 55 cm D x 72 cm H) and built-in automation to provide ease of operation.

### Automation from Setup to Analysis

The SH800S provides the highest level of automation and ease of use available in a cell sorter. Researchers can accurately set up, calibrate, and monitor sorting with a push of a button. This delivers true ease of operation to save time and improve consistency of results.

A novel microfluidic sorting chip uses patented CoreFinder™ technology to automate key steps of instrument setup and operation, streamlining workflow.

The system is versatile, allowing researchers to sort of a wide range of cell sizes to accommodate application requirements. Chip sizes include 70  $\mu$ m, 100  $\mu$ m, and 130  $\mu$ m.



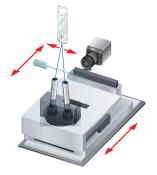
Sample Collection Slide Door

#### Automated Chip Loading and Positioning

System setup begins with a one-touch installation to load the microfluidic flow cell chip. Actuators ensure precise positioning of the chip inside the chip loader. Sheath, sample, and vacuum lines connect and seal automatically to their respective ports once the chip is loaded. Made of durable plastic, the chip is easy to replace when needed.

# Automated Side Stream Calibration

The angle and the position of the side streams are calculated and adjusted during setup for tube and plate sorting. This ensures that the sort stream is centered in the collection tube automatically without manual adjustment.

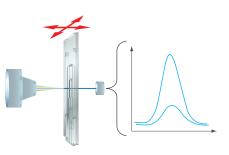


#### Automated Optical Axis Adjustment

The alignment of the chip to the lasers is optimized automatically during setup using the Sony patented Blu-ray<sup>M</sup> technology for aligning and tracking laser position. On a daily basis, using AutoSetup beads, the X and Z positions of the chip are adjusted to ensure consistent results.

#### Automated Droplet Calibration

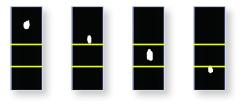
The droplets are automatically calibrated by adjusting the frequency and the drop drive to achieve an optimal breakoff point (BOP) for each type of sorting chip.





#### Automated Drop Delay Calibration

A dedicated laser and camera perform real-time analysis of droplet images using AutoSetup beads. Patented technology calculates drop delay by capturing the fluorescence image of the drop-delay beads. The relationship between bead positions and drop delay is used to calculate the optimal drop delay to enable precise targeting and high yield. Automatic analysis of the binary image of the droplets, in real time, calculates the drop delay using a patented algorithm.



#### Automated Sort Monitoring

The SH800S software monitors and actively makes adjustments to the drop drive to maintain a stable breakoff point. This feature ensures consistently good sort performance and allows detection of clogs, and empty tubes, and facilitates walk-away operation.



### Software

Software, sensors, and CoreFinder patented technology provide automation across the workflow from setup to shutdown to ensure consistency, save time, and improve the accuracy of results. The interface is designed with ribbon tabs that logically organize features to make them rapidly accessible. An experiment-centric approach makes the software easy to teach, learn, and use.

Software wizards include step-wise workflows that guide users through startup, multicolor compensation, sorting, and system cleaning. Since the system simplifies even advanced sorting techniques, it can be used by researchers with little sorting experience.

#### System Startup

Upon startup, the system initialization includes diagnostics that ensure all subsystems are properly working. Once verified, the system status and a green ready message are displayed on the LCD monitor on the front of the instrument.



The LCD monitor on the front of the SH800S displays status information during operation.

#### Initial Instrument Setup, Precision Alignment, and Calibrations

The setup wizard guides researchers through the process of loading a sorting chip, selecting lasers, and inserting the optical filters required.

#### Fluidics Check

When complete, the fluidics check starts, and sheath fluid droplets appear from the tip of the sample probe. If needed, debubble and sample line cleaning wizards can be run. Once complete, the auto calibration screen is displayed.

#### Alignment and Calibrations

The system automatically and precisely aligns the sorting chip to the lasers, calibrates the droplets and side streams, and estimates the drop delay using AutoSetup beads.



Automatic calibration performs precise measurements using the side stream monitor. The software displays progress and status at each stage of calibration.



The QR code on the packaging is scanned to identify the chip information, including nozzle size.



The sorting chip is easy to load.



To further simplify setup, the Fluorochrome Detection Matrix screen displays where to insert optical filters.

#### **Experiment Settings**

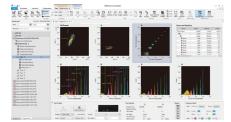
Users can create an experiment by selecting a new template, a recent experiment, or a shared template (public) from the Experiment window. If a new template is selected, dialogs guide the user to choose experiment settings such as sample groups, tubes, and pulse parameters for data acquisition. Once a template is selected, the user can start acquisition or the compensation wizard.



Selecting a template or a recent experiment on the left of the window displays the experiment structure on the right.

#### Data Analysis and Display

Data is displayed as dot plots and histograms on worksheets, and events can be marked using gates. The software has a number of tools to select, adjust, label, and measure statistics of target populations. Data can be easily exported in FCS formats (3.0 or 3.1) to use with third party analysis software.

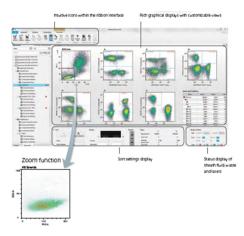


The Experiments tab provides easy access to all lists of previous experiments as well as visualization and statistics tools for analysis.

#### Sorting

Once events are gated on plots, targeted cells can be sorted for further analysis. Sort control is managed in a simple dialog where settings and configurations are selected. The SH800S supports a variety of sorting methods and vessels including 2-way tubes, 8-well strips, slides, and 6-, 12-, 24-, 48-, 96-, and 384-well plates.

The system supports eight sorting modes to provide different levels of purity (including single-cell mode) and yield to enable optimal results. In addition, the software provides an assistive mode for large cells to improve sorting performance. Three sorting chip nozzle sizes (70  $\mu$ m, 100  $\mu$ m, and 130  $\mu$ m) are available to accommodate a range of cell sizes and applications.



Sorting settings and sort status are easily accessed and customized.

#### System Maintenance

The SH800S software uses wizards to guide users to perform routine tasks such as fluidics cleaning for aseptic sorting and parts maintenance.

## Options

#### **Biosafety Cabinet**

The BCC300AMS Class II biosafety cabinet custom designed for the SH800S by The Baker Company is available as an option to provide personnel and product protection. The cabinet measures 118 cm (W) x 99.1 cm (D) x 224 cm (H).

**Biosafety Standards Compliance** The cabinet was tested by The Baker Company using microbiological assays with the SH800S sorter inside the work area. The testing concluded that the biosafety cabinet with the SH800S inside met several international biosafety standards including the National Sanitation Foundation Standard 49 (NSF49) and the European Standard 12469.

#### **Built-in Aerosol Management**

The cabinet incorporates a built-in aerosol management system which operates independently to actively evacuate aerosols from the sort collection chamber. The dual routes of aerosol evacuation ensure maximum personnel protection.



#### E-Beam Sterilized Consumables

For applications in which cross contamination between samples or external biological entities are a factor, optional e-beam sterilized consumables including chips, setup beads, and sample lines can be used to streamline workflow.



Sort Deposition System

The optional Sort Deposition System facilitates high-throughput sorting and precise deposition of cells into 6-, 12-, 24-, 48-, 96-, and 384-well plates or PCR plates.



96-well plate holder

#### **Exchangeable Sample Fluidics**

The SH800S sorting chip is an integrated flow cell/nozzle assembly. It contains microfluidic channels for controlling the flow of sample and sheath fluid. Within the chip, the sample is interrogated by the lasers before it passes through the nozzle for sorting.

#### **Easy Installation**

Chip installation and removal are quick and easy to reduce the downtime associated with changing nozzles during setup and removal of clogs. The chip and the PEEK sample line-chip connector assembly, which come in contact with the sample, are fully disposable. This gives researchers the option to change out the sample fluidics path if needed.

#### Index Sorting Software

Index sorting software records the X and Y coordinates of each event sorted into a multiwell device. This feature allows researchers to track the scatter and fluorescence intensity of individual cells sorted in each well.

#### Gates and Statistics

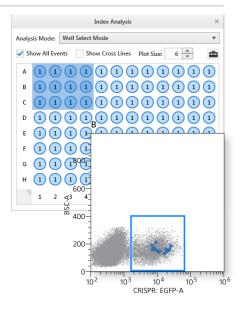
Name	Events	%Parent	%Total
All Events	96	0.00%	100.00%
A	95	98.96%	98.96%
B B	95	100.00%	98.96%
EGFP+	95	100.00%	98.96%

Single cells sorted into a 96-well plate.

#### Versatile Application Support

The SH800S offers 70- $\mu$ m, 100- $\mu$ m, and 130- $\mu$ m microfluidic sorting chips to permit the sorting of a wide range of cell sizes and applications.

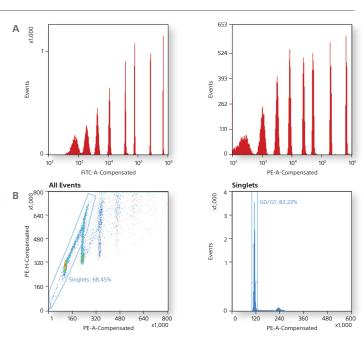




### Applications

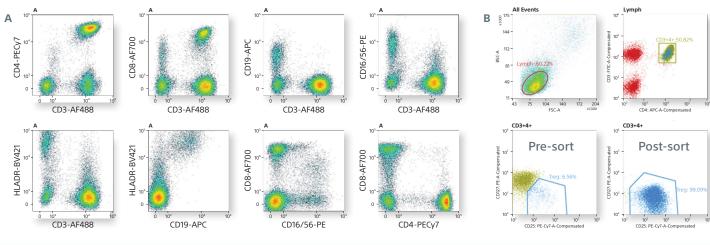
#### **Resolution and Sensitivity**

A. Fluorescence sensitivity MESF values measured using Spherotech<sup>®</sup> 8-peak beads are: FITC 120 and PE 110. B. Fluorescence resolution was measured using propidium iodide (PI) stained chicken erythrocyte nuclei (CEN). A coefficient of variation of <2.5% was observed for the GO/G1 peak.



#### Immunophenotyping Assays

Distinct resolution of multicolor samples is seen with a four-laser SH800S system. A. Normal human blood was stained with CD3 Alexa Fluor® 488, CD4 PE-Cy™7, CD8 Alexa Fluor® 700, CD19 APC, CD16+CD56 PE, and HLA-DR BV421. All plots were gated on lymphocytes. B. High-purity sorting of the regulatory T cells (CD3+ CD4+ CD25 high CD127-) population.

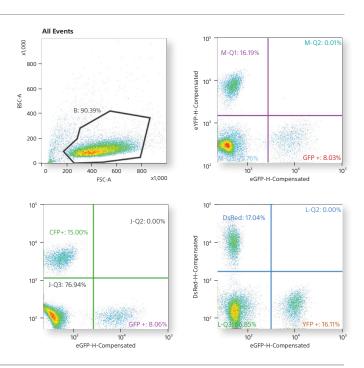


#### Gates and Statistics

Name	Events	%Parent	%Total
<ul> <li>All Events</li> </ul>	30,000	0.00%	100%
Lymph	18,067	60.22%	60.22%
• CD3+4+	9,181	50.82%	30.60%
<ul> <li>Lymph</li> </ul>	602	6.56%	2.01%

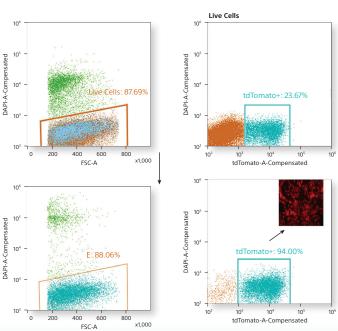
#### Fluorescent Protein Analysis

Human cell lines co-expressing GFP, YFP, dsRed, and CFP using the fluorescent protein optical filters are shown. Refer to the filter guides for the fluorescent protein filter sets.



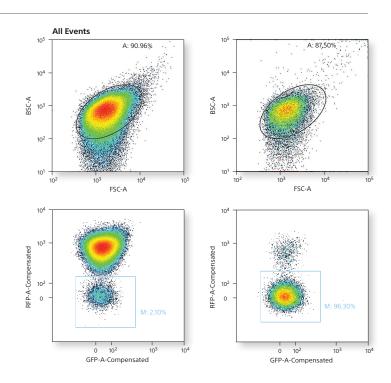
#### Sorting and Viability Analysis

These plots show a sorting and viability analysis of differentiating mouse embryonic stem cells expressing IsI1Cre-tdTomato. Analysis of the tdTomato (+) purified population is shown. The inset (lower right plot) shows an image of cells in culture 24 hours post sorting.



#### **Small Particle Resolution**

High speed sorting of *E.coli* using a 70- $\mu$ m sorting chip. A concentrated sample of *E.coli* cells was sorted at a threshold event rate of 30,000 events/s at a sort setting of 50 kHz and 40 psi. Postsort analysis shows that >95% purity was obtained.



### Filter Guide

#### Fluorochrome Guide

Filter Set 1	FL1	FL2	FL3	FL4	FL5	FL6
EGFP	•					
FITC	•					
Alexa Fluor® 488	•					
EYFP	•					
mCitrine	•					
CFSE	•					
PE		•				
PE-Texas Red®			•			
Propidium lodide			•			
dsRed			•			
tdTomato			•			
mCherry			•			
mPlum				•		
7-AAD				•		
PE-Cy™5				•		
PerCP				•		
PE-Cy™5.5					•	
PerCP-Cy5.5					•	
PE-Cy7						•
APC				•		
Cy5				•		
Alexa Fluor <sup>®</sup> 647				•		
APC-Cy5.5					•	
Alexa Fluor <sup>®</sup> 700					•	
APC-Cy7						•
APC-Alexa Fluor® 750						•

Filter Set 2	FL1	FL2	FL3	FL4	FL5	FL6
BD Horizon Brilliant™ Violet	•					
Alexa Fluor <sup>®</sup> 405						
DAPI						
Pacific Blue™	٠					
mCFP	٠					
Hoechst 33342	٠					
EGFP		•				
FITC		•				
Alexa Fluor <sup>®</sup> 488		•				
EYFP		•				
mCitrine		•				
CFSE		•				
PE			•			
PE-Texas Red®			•			
PE-Dazzle™			•			
Propidium lodide			•			
dsRed			•			
tdTomato			•			
mCherry			•			
mPlum				•		
7-AAD				•		
PE-Cy5				•		
PE-Cy5.5					•	
PerCP-Cy5.5					•	
PE-Cy7						•
APC				•		
Alexa Fluor® 647				•		
APC-Cy5.5					•	
Alexa Fluor® 700					•	
APC-Cy7						•
APC-Alexa Fluor® 750						•

### Filter Set 1 includes:

LP1 639LP	FL1 525/50	FSC 488/17F 488/17F
LP2 600LP	FL2 585/30	DCC 400 (47D 400 (47D
LP3 561LP	FL3 617/30	BSC 488/17B 488/17B
LP4 752LP	FL4 665/30	
LP5 685LP	FL5 720/60	
	FL6 785/60	

#### Filter Set 2 includes:

LP1 639LP	FL1 450/50	FSC 488/17F
LP2 561LP	FL2 525/50	DCC 400 (47D
LP3 487.5LP	FL3 600/60	BSC 488/17B
LP4 752LP	FL4 665/30	
LP5 685LP	FL5 720/60	
	FL6 785/60	

# Specifications

	Excitation lasers	488 nm, 405 nm, 638 nm, 561 nm
	Output power	30 mW (max.) optical fiber output
o .::	Beam alignment	Collinear optical system
Optics	Detection parameters	6 fluorescence + 2 scatter
	Analog-to-digital converters (ADC)	8-channel 20-bit, 110 MHz
	Pulse measurement	Height, Area, Width
	Sample tube	Single, auto-loading tube
	Tube types	0.5-mL, 1.5-mL, 5-mL, and 15-mL tubes
	Sort devices	2-way tube, multiwell plates, PCR tubes, slides
Fluidics	Temperature control	5°C, 37°C (electric cooling method)
	Agitation unit	Eccentric rotation
	Magnetic drive	300-rpm speed
	Sorting chip size	70 µm, 100 µm, 130 µm
	Event rate	100,000 eps
Sort Performance	Sorting speed	Using the 70-µm sorting chip at 50 kHz, an average threshold of 12,000 events per second can be achieved with >98% purity and >80% yield. A threshold rate up to 30,000 events per second can be achieved without affecting purity but with a decrease in yield based on Poisson's statistics.
renormance	Scatter resolution	0.5 μm
	Fluorescence resolution	<2.5% half-peak coefficient of variation (HPCV)
	Fluorescence sensitivity	FITC 120 MESF, PE 110 MESF
	Dimensions	W: 21.7 in. (55 cm) x D: 21.7 in. (55 cm) x H: 28.4 in. (72 cm)
	Fluidics cart	W: 30 in. (76.2 cm) x 20.5 in (52.1 cm) x 22.8 in (58.0 cm)
	Weight	216 lb (98 kg)
	Fluidics cart	71 lb (32 kg) (dry weight)
Ancillary	LCD panel	7-inch, 800 x 480 pixels
	Power supply	100-240 V, 50/60 Hz
	Power consumption	500 W (max.)
	Operating temperature	17.5°C to 27.5°C
	Relative humidity	20% to 80%
Compliance:	Operating system	Microsoft® Windows® 8 Professional, 64 bit
Environmental Rohs	Data file structure	Flow Cytometry Standard (FCS) 3.0 or 3.1
	Safety standards compliance	UL, CE, CSA





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