Cytek® Aurora CS

Where Cellular Discoveries Take Focus

CYTEK®

TRANSCEND THE CONVENTIONAL

Meet Aurora CS

A prodigy built from the Cytek® Aurora system's unique combination of innovative technologies that takes cell sorting to the next level of performance and flexibility.

With up to five lasers, three scattering channels, and 64 fluorescence channels, the Aurora CS system is designed to leverage the paradigm-shifting technology onboard the Cytek Aurora system and deliver the sorting capabilities you have come to expect in a high-end sorter.

Like the Aurora system, the Aurora CS system provides the benefits of Full Spectrum Profiling™ (FSP™) technology. Its optical design and unmixing algorithm provide scientists increased flexibility, enabling the use of a wide array of new fluorochrome combinations without reconfiguring the system for each application. The state-of-the-art optics and low-noise electronics provide high resolution, high content and high sensitivity.

The result is a system that delivers high resolution at the single cell level to resolve the most challenging cell populations, such as cells with high autofluorescence or low levels of expression of key biomarkers, regardless of assay complexity, and to isolate live cells for downstream studies. Welcome to a new world where the assays optimized on the Aurora can also be run on the Aurora CS system for sorting.

As a sorter, the Aurora CS system offers the flexibility required to meet various biological and sorting conditions. With up to 6-way sorting, customizable nozzle settings and sort modes, automated drop delay and sort stream monitoring, the Aurora CS system offers the flexibility required to meet various application needs from smaller to larger cells.

SpectroFlo® CS software is used to drive the Aurora CS system. Its sorter UI console offers intuitive workflows combined with powerful and customizable functionality.

In summary, the Aurora CS system enables you to take your research even further, gain deeper insights, and accelerate your pace of discovery.

See more. Sort more.

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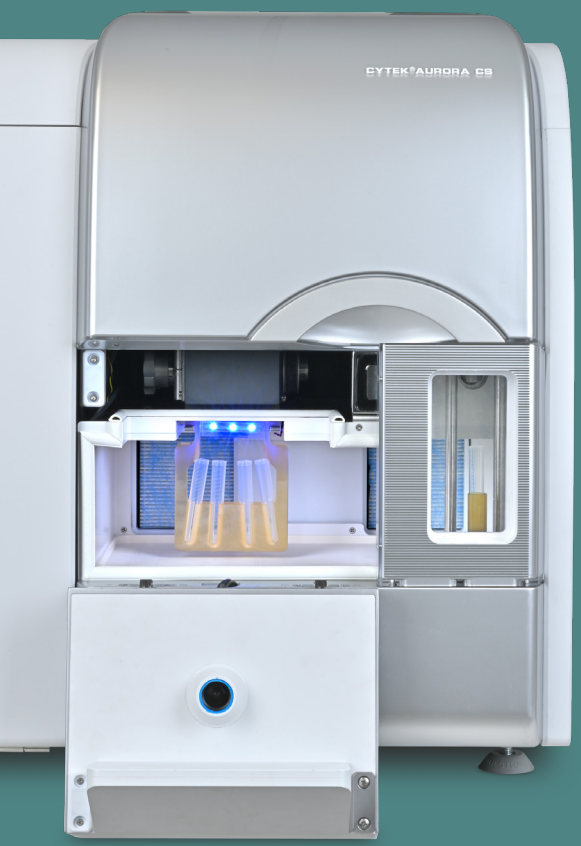
Built with Cytek's Revolutionary Technologies:
Providing Excellent Single Cell Resolution

The Aurora CS system is capable of up to 67 detection channels (64 fluorescence channels, FSC, blue laser SSC, and violet laser SSC) and is empowered by revolutionary technologies, including:

- Proprietary high sensitivity Coarse Wavelength Division Multiplexing (CWDM) semiconductor detector arrays enable efficient spectrum capture for dyes emitting in the 365-829 nm range.
- High bandwidth electronics design scalable up to 67 channels.
- Integrated HEPA filtration system as a first layer of protection from aerosols.
- Magnetically controlled plate sorting and smooth, easy to clean surfaces.



Why Choose the Cytek® Aurora CS?

So Many Colors

40 colors demonstrated including fluorochromes with emission spectra in close proximity to each other.

Exceptional Sensitivity and Resolution

Sensitivity redefined using state-of-the-art optics and low-noise electronics.

Extract autofluorescence and improve resolution of highly autofluorescent samples.

New Levels of Flexibility

No need to reconfigure optical filters for different fluorochromes.

Use any commercially available fluorochrome excited by the onboard lasers.

Choose from a variety of sample input and collection devices including 5 and 15 mL tubes for input and 96-well plates, 1.5 and 5 mL tubes for collection.

Seamless Sorting Experience

Automated drop delay, sort monitoring, and clog detection for a reliable sorting experience.

Comprehensive sort reports automatically record settings used from every sort.

Assay transferability from the Cytek Aurora system or conventional flow cytometers.

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Feature Highlights

The Cytek® Aurora CS system is designed for sorting performance and application flexibility, beginning with sort set-up and carrying through to the sorted sample output. Here are just a few examples. See these and more by requesting a demo with your Cytek technical sales representative (sales@cytekbio.com).

Sample Input Flexibility and Control

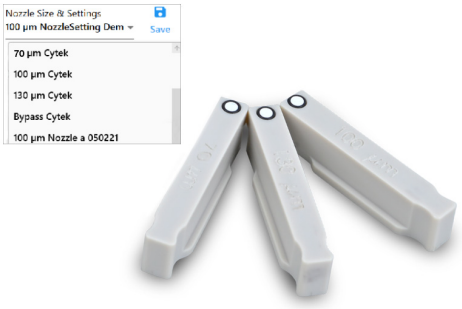
The Cytek Aurora CS system's sample loading chamber accommodates 5 and 15 mL polystyrene or polypropylene tubes. In the SpectroFlo® CS software user interface, the following options are available:

- Toggle on the chamber light to see how much sample remains in the tube
- Choose from three sample tube mixing speeds
- Set the chamber temperature to cool or heat your sample input and sort collection tubes
- After sorting, backflush remaining sample from the sample line into the sample tube to sort as many cells as possible

Variety of Nozzle Settings

The ideal nozzle diameter for an optimal sort output depends on the size and fragility of the particles of interest. Choose from 70, 100, and 130 µm* default nozzle settings or customize and save your own setting inside SpectroFlo® CS software for future reuse.

*130 µm available on request



Predefined and Custom Sort Modes

Select one of Cytek's predefined sort modes or create a custom defined sort mode to meet the needs of each user's sorting application.

Purity

Isolate the population of interest with little to no contaminants from other populations

Enrich

Prioritize retrieving a high number of the target population with reduced sort purity

Multiway

Intended for 4- or 6-way sorting for efficient drop deflection

Single Cell

Isolate single cells into 96-well plates

Mixed

A combination of Purity and Enrich modes

Custom

Adjust the sort decision settings to meet your application needs

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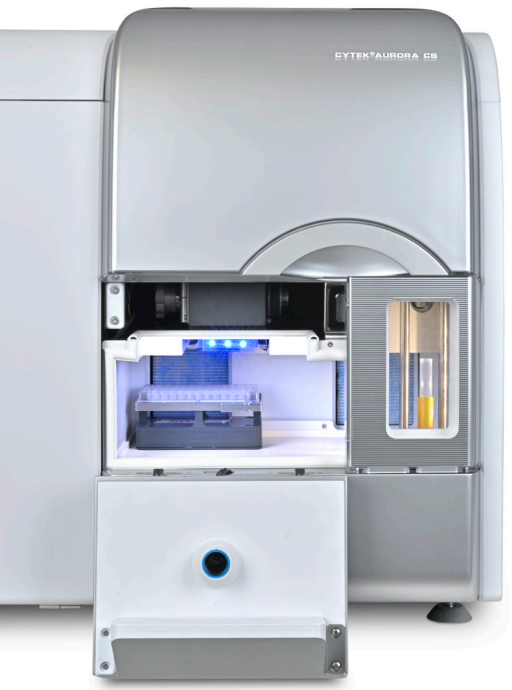
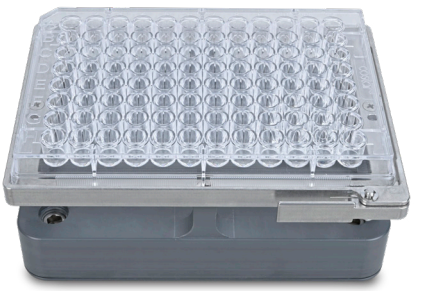


Plate Sorting

Sort into 96-well plates by defining the sort settings for each well inside SpectroFlo® CS software, loading a plate into the plate holder, and placing the holder inside the droplet deposition unit.

Using the plate sorting high throughput mode, deposit 1 cell into 96 wells in less than 2 minutes. Utilize the index sorting feature to track and overlay data from each plate well in the software's analysis plots. Additionally, use the detailed sort reports for a comprehensive record of the sort set-up and performance for each experiment.



Multi-Way Sorting

Sort up to 6 ways into 1.5 mL tubes, or up to 4 ways into 5 mL tubes. Easily glide the tube holders into the droplet deposition unit, and use integrated live video feed and stream aiming tools inside SpectroFlo® CS software to adjust sort streams.

Biosafety Controls

Primary aerosol evacuation is integrated into the Aurora CS system with user-replaceable HEPA filters. The timers built into SpectroFlo® CS software remind users when to replace the filters.

SpectroFlo® CS software displays instrument indicators for warm up, HEPA filter replacement, remaining sheath, waste tank level, and connection.

For an optional secondary level of protection, add on a small footprint Biosafety Class 2 Type A2 cabinet. The cabinet comfortably houses the Aurora CS system with room to the right of the instrument for a sample tube rack and other small commonly used laboratory items.

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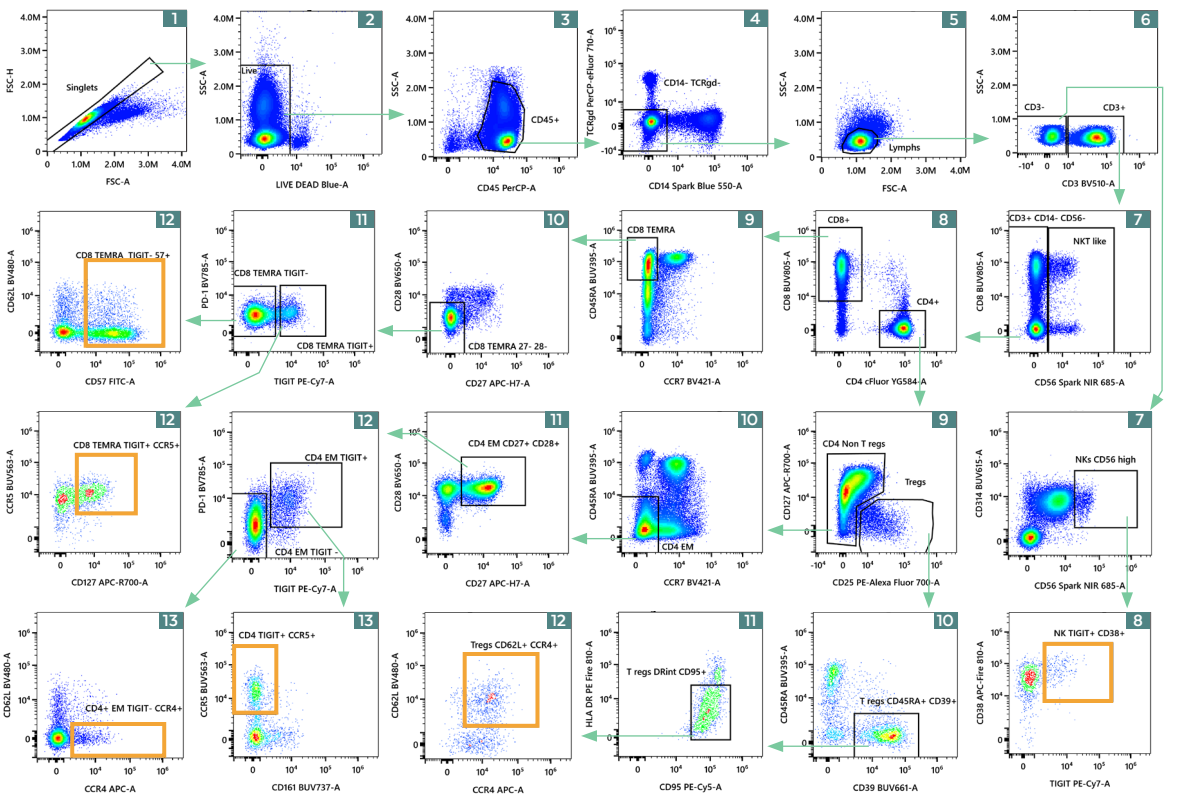
In-Depth 6-Way Sorting

The Cytek® Aurora CS system combines Full Spectrum Profiling™ (FSP™) technology and high-end sorting functionality to enable you to dig deep into your cellular assays. To demonstrate the Aurora CS system's capabilities, made possible by its advanced electronics design, a 28-color human deep T-cell and NK cell immunophenotyping panel was transferred from the Cytek Aurora. Six low frequency T-cell and NK cell subsets shown below were sorted with a 100 µm nozzle and checked for purity.

28-Color Human Deep T-Cell and NK Cell Immunophenotyping Panel

UV	Violet	Blue	Yellow-Green	Red
BUV395 CD45RA	**BV421** CCR7	**FITC** CD57	**cFluor® YG584** CD4	**APC** CCR4
LIVE/DEAD Blue	**BV480** CD62L	**Spark Blue 550** CD14	**PE/Dazzle 594** CD337	**Spark NIR 685** CD56
BUV563 CCR5	**BV510** CD3	**PerCP** CD45	**PE-Cy5** CD95	**APC-R700** CD127
BUV615 CD314	**BV650** CD28	**PerCP-eFluor 710** TCRγδ	**PE-Alexa Fluor 700** CD25	**APC-H7** CD27
BUV661 CD39	**BV750** CXCR5		**PE-Cy7** TIGIT	**APC/Fire 810** CD38
BUV737 CD161	**BV785** PD-1		**PE/Fire 810** HLA-DR	
BUV805 CD8				

Sort Populations Defined Deep Into the Gating Hierarchy



The sort gates for each of the six sorted populations are shown in orange in the gating strategy above. The frequency of each sorted population ranged from 0.09 to 0.40% of total cells in the sample. The level in the gating hierarchy for each sort gate is shown by the green rectangle in the upper right corner of each pseudocolor plot, going as deep as 13 levels into the hierarchy.

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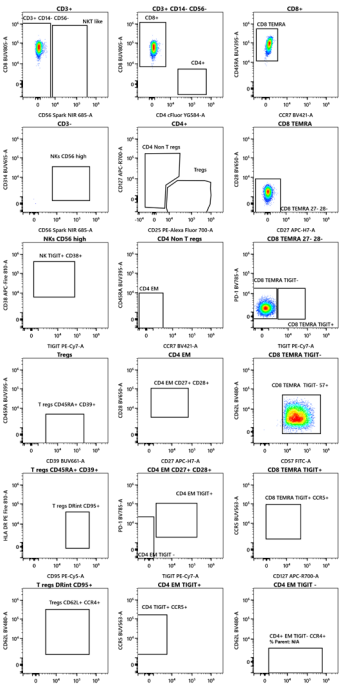
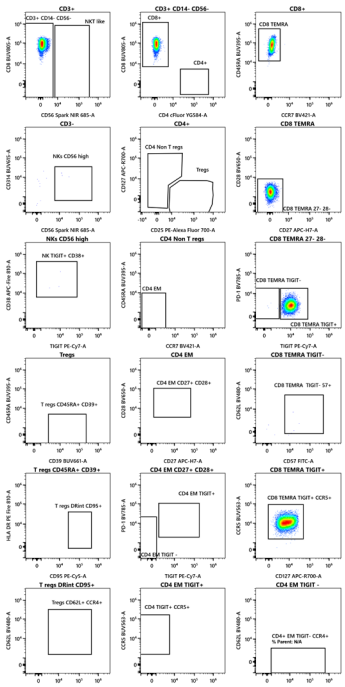
Sort Purity Checks

The plots below show the post-sort purity results for each of the six sorted populations.

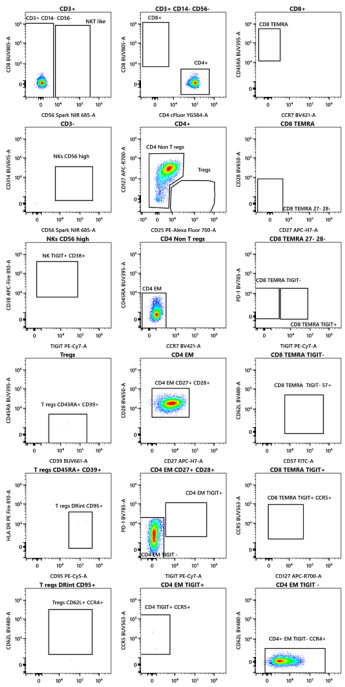
CD8⁺ TEMRA TIGIT⁻ CD57⁺
Purity: 99.5%



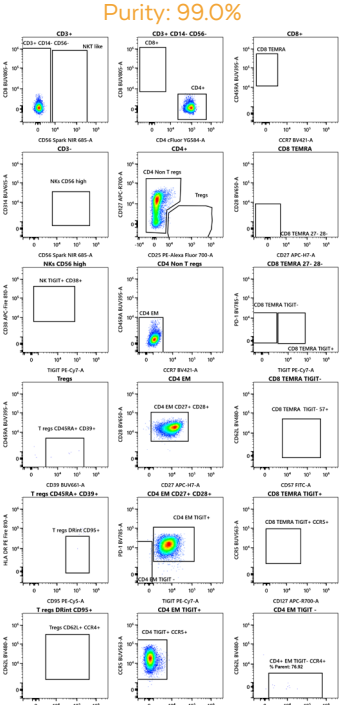
CD8⁺ TEMRA TIGIT⁺ CCR5⁺
Purity: 99.7%



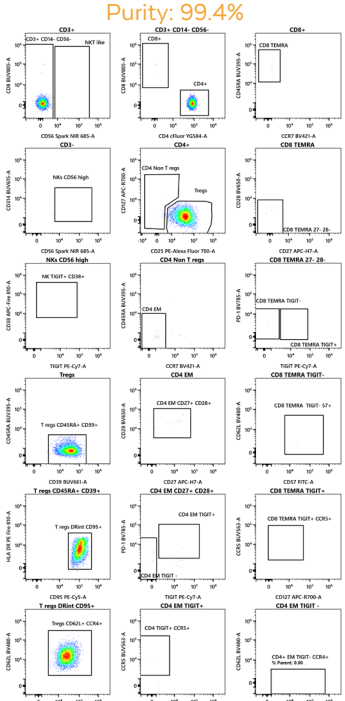
CD4⁺ EM TIGIT⁻ CCR4⁺
Purity: 99.2%



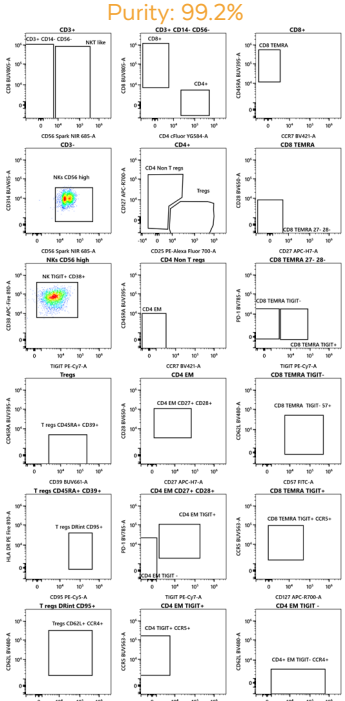
CD4⁺ TIGIT⁺ CCR5⁺
Purity: 99.0%



Tregs CD62L⁺ CCR4⁺
Purity: 99.4%



NK TIGIT⁺ CD38⁺
Purity: 99.2%



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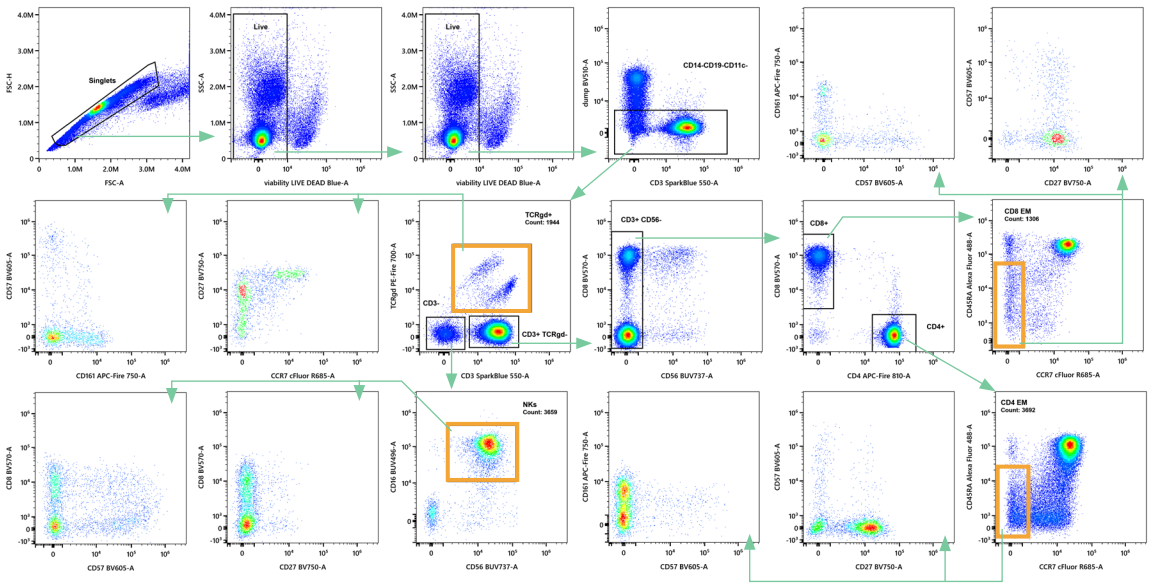
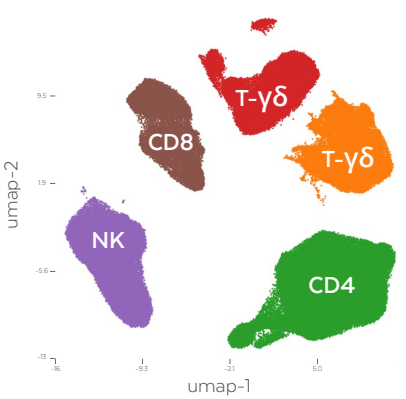
Take Sorted Cells Into Functional Assays

When sorting cells for downstream experimental assays, it is crucial that the sorted cells remain viable and functional post-sorting. To demonstrate this capability with the Cytek® Aurora CS system, human PBMCs were prepared and sorted 4-ways with a 70 µm nozzle to collect CD4+ effector memory, CD8+ effector memory, TCRγδ T cells, and NK cells using the gating strategy below. Sort gates are outlined in orange.

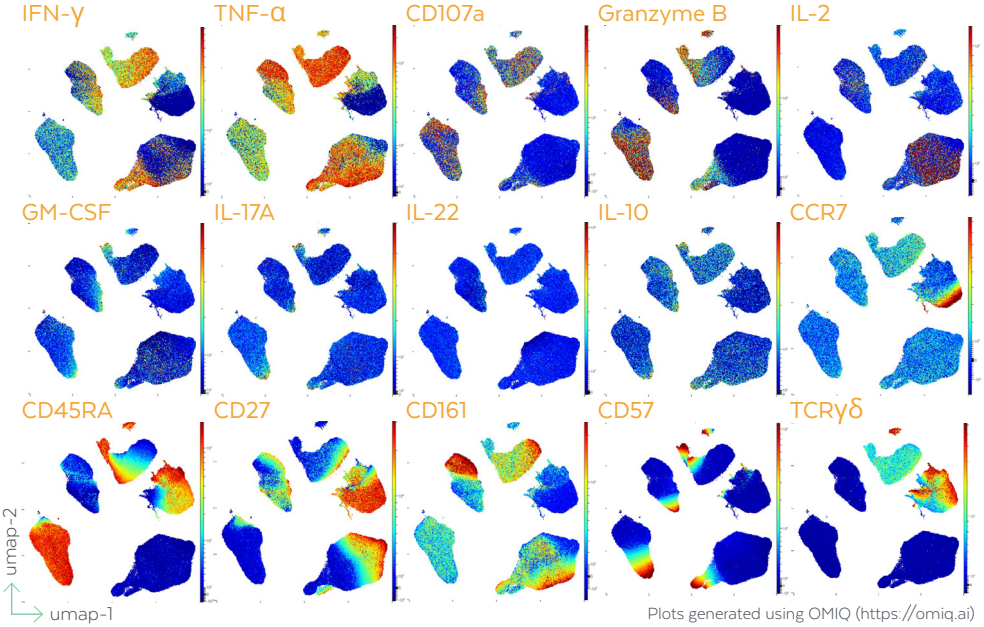
After sorting was completed, the cells were rested for 2 hours in an incubator, then stimulated with PMA and ionomycin in the presence of BFA, monensin, and CD107a, and next stained with 8 different intracellular markers. The results are shown in the high dimensional unsupervised analysis figures below generated using OMIQ and the 2-D plots on the next page generated using SpectroFlo® CS software.



UMAP Plots of FlowSOM Clustered Post-Sort Stimulated Cell Results



Single live cells from every sorted population were clustered together using FlowSOM and UMAP for dimensionality reduction and cluster visualization. The scatter plot above shows the 5 main clusters identified. Expression of activation markers is shown in colored-continuous scatter plots. The 2 TCRγδ clusters identified show a distinct and opposite pattern of activation in term of production of IFN-γ, TNF-α, CD107a, and granzyme B.



Plots generated using OMIQ (https://omiq.ai)

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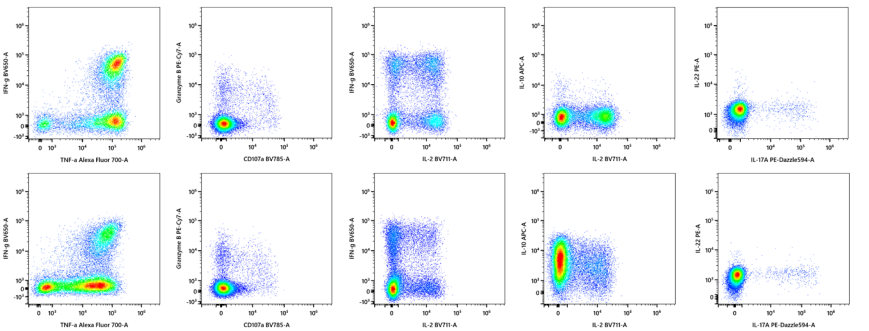
2-D Plots Post-Sort Cell Functionality Results After PMA/Ionomycin Stimulation

CD4+ Effector Memory Cells

Sorted

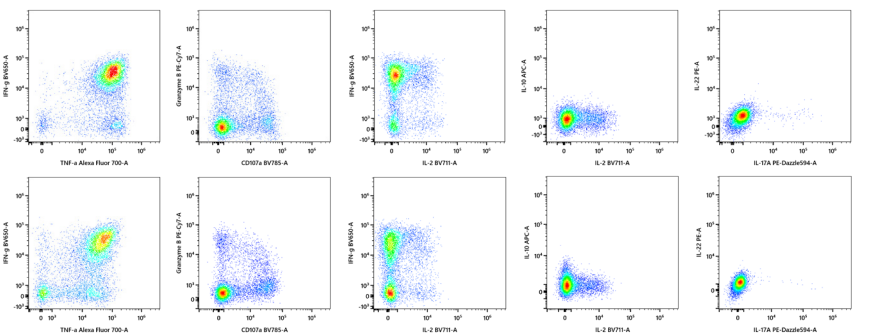
Unsorted



CD8+ Effector Memory Cells

Sorted

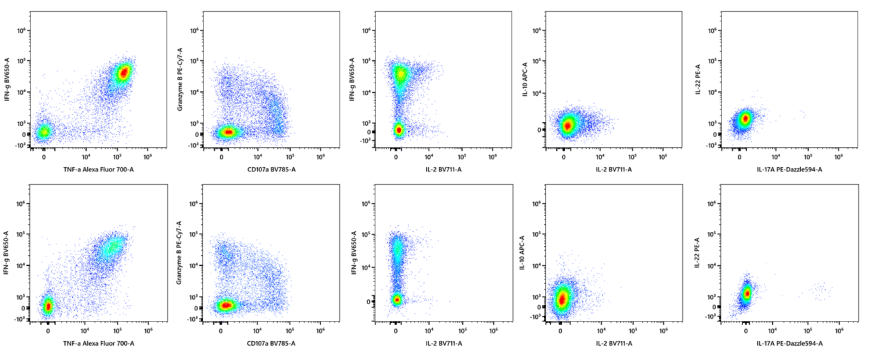
Unsorted



TCRγδ T Cells

Sorted

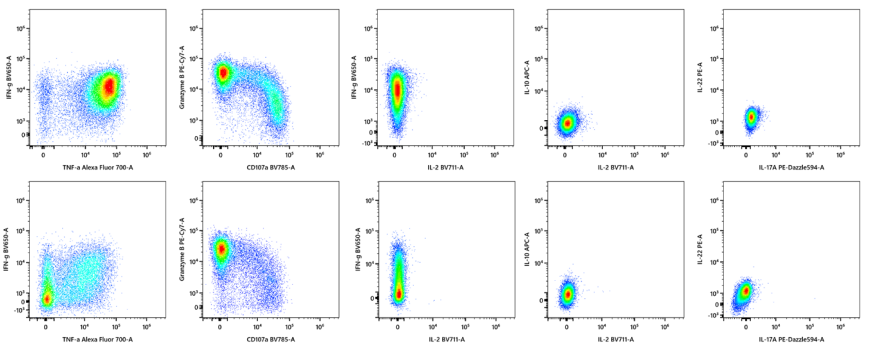
Unsorted



NK Cells

Sorted

Unsorted



Unsorted cells and cells from each of the 4 sorted populations were stimulated with PMA and ionomycin in the presence of BFA, monensin, and CD107a. After stimulation, cells were stained with 8 different intracellular markers. The cells were acquired on a Cytek Aurora system, and the results displayed using 2-D plots in SpectroFlo® CS to visualize cell CD107a expression and cytokine production patterns. The cell numbers for each population in the unsorted or sorted populations are different due to the nature of this experiment.

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Resolving Challenging Dye Combinations

The detection of some fluorochrome combinations by conventional flow cytometry presents a challenge due to high amounts of spectral overlap (Figure 1, 4). The Cytek® Aurora CS system addresses this challenge by using Full Spectrum Profiling™ (FSP™) to detect differences in full emission spectra signatures across all lasers to clearly resolve these combinations, even if the populations are co-expressed (Figures 2, 3, 5, and 6).

Example 1: APC and Alexa Fluor 647

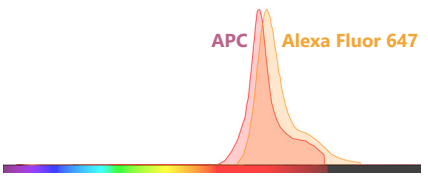

Figure 1: Spectrum plots from a conventional spectrum viewer shows heavy overlap between APC and Alexa Fluor 647.

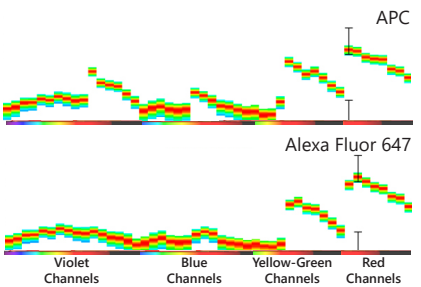

Figure 2: Spectrum plots from a four-laser Aurora system show distinct signatures for APC and Alexa Fluor 647.

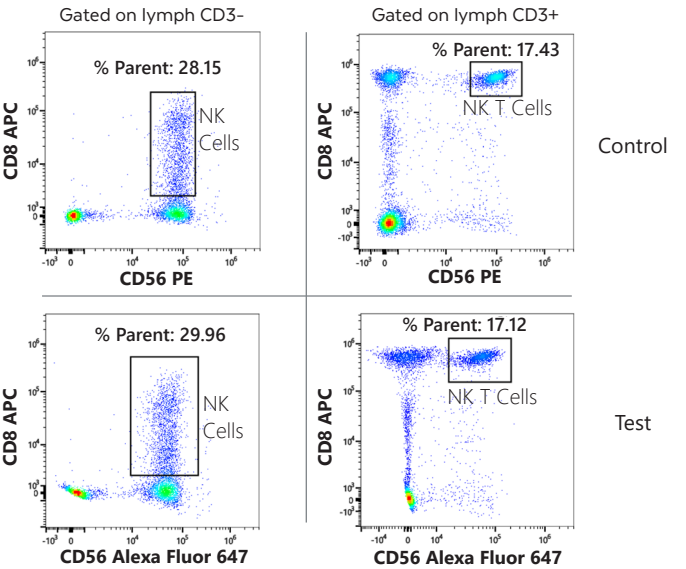

Figure 3: Whole blood from a healthy donor was stained, lysed, washed, and analyzed on a four-laser Aurora system. Subsets of NK and NK T cells that co-express CD56 Alexa Fluor 647 and CD8 APC were easily identified. For comparison, blood from the same donor was stained with CD56 PE and CD8 APC and yielded similar percentages of NK and NK T cells, demonstrating that APC and Alexa Fluor 647 combined did not impact results.

Example 2: BFP, GFP, and mCherry

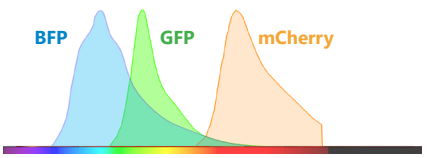

Figure 4: Spectrum plots from a conventional spectrum viewer.

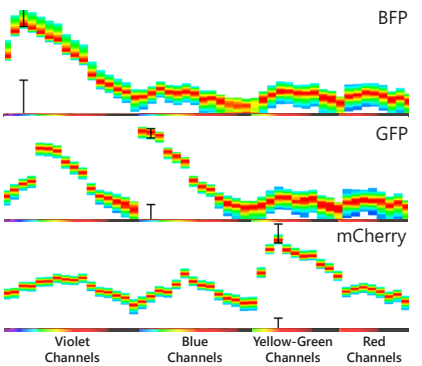

Figure 5: Spectrum plots from a four-laser Aurora system show distinct signatures for BFP, GFP and mCherry.

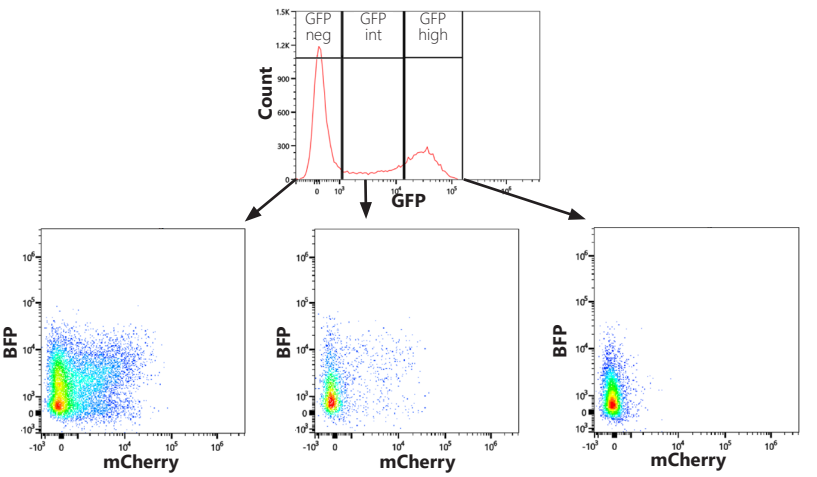

Figure 6: AB2.2 mouse embryonic stem cells were genetically modified to stably express BFP, GFP and mCherry under the control of different fate marker promoters. The stable cell line generated was then cultured under differentiation conditions, harvested, and analyzed on a four-laser Aurora system to assess the expression of fluorescent proteins. Autofluorescence extraction was used to enhance results. Sample courtesy from Luigi Russo, Hannah L. Sladitschek and Pierre Neveu, Cell Biology & Biophysics, Neveu group, EMBL.

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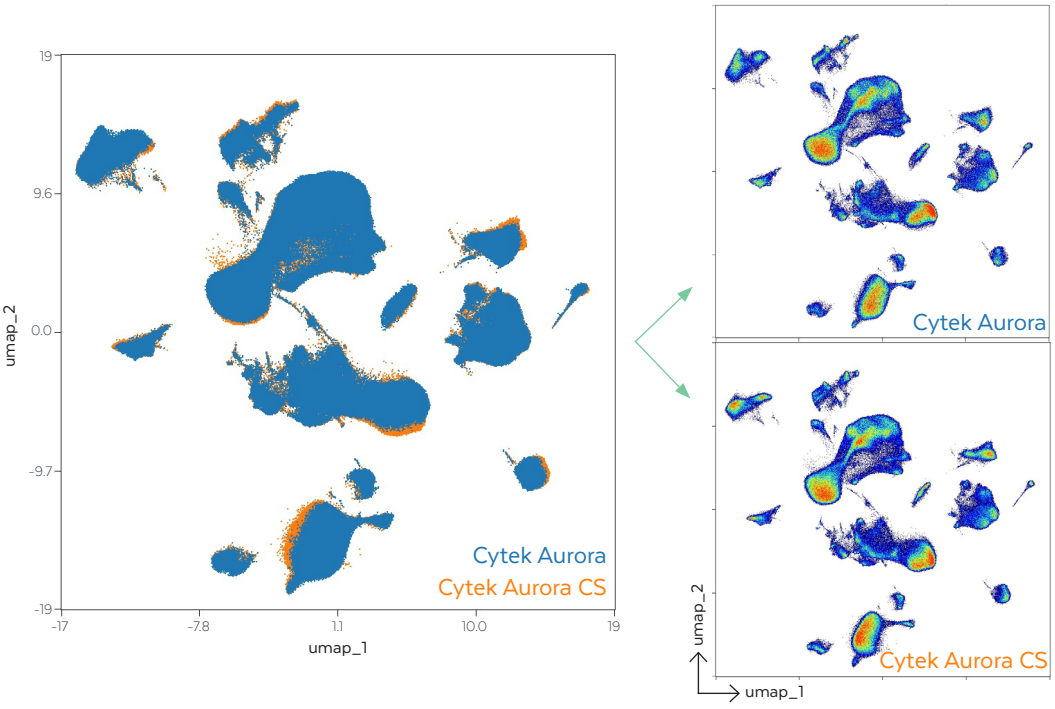
Excellent Optical Performance

With Full Spectrum Profiling™ (FSP™) Technology

The ability to generate high quality multi-parametric data is key to successfully utilize data dimensionality and 2-D visualization algorithms. Cytek's patented FSP™ technology is built into both the Cytek Aurora system and the Cytek Aurora CS system so users of both can generate high quality data. In OMIP-069 (PMID: 32830910), Cytek's 40-color deep immunophenotyping panel for the Cytek Aurora can perform equally well on the sorting-capable Cytek Aurora CS system.

To demonstrate this:

1. 40-color human PBMC samples were prepared.
2. Reference controls and samples were acquired on both the Cytek Aurora system and Cytek Aurora CS system.
3. Data from both systems was gated on live singlet CD45^{+} lymphocyte and monocyte populations and visualized in 2-D using UMAP.
4. A UMAP scatter plot of the data from each system is shown overlaid below with a high level of comparability. Note the Aurora system (blue) and Aurora CS system (orange) follow the same patterns and almost fully overlap.
5. UMAP density plots of the data from each system are shown side-by-side.



Have you read OMIP-069? Download it now from the resources publication collection at cytekbio.com.

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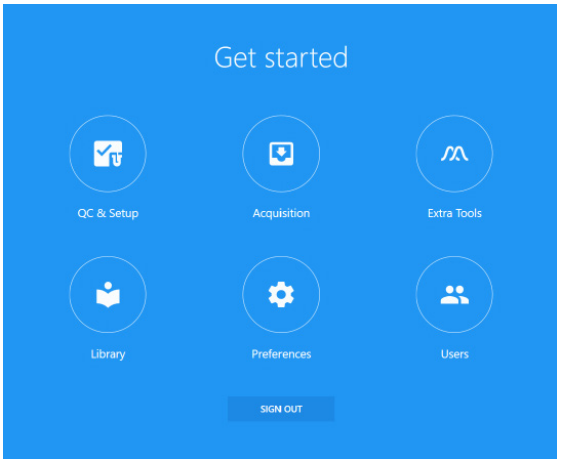
SpectroFlo® CS Software Acquisition Workflows

SpectroFlo® CS software offers an intuitive workflow from quality control (QC) to data analysis with technology-enabling tools that simplify running any application.

QC and Setup:
Run Daily QC to monitor instrument performance and add reference controls.

Library:
Add or remove experiment templates, worksheet templates, fluorochrome information, QC bead information, and more.



Extra Tools:
Unmix data using controls from different experiments or apply virtual filters to your data.

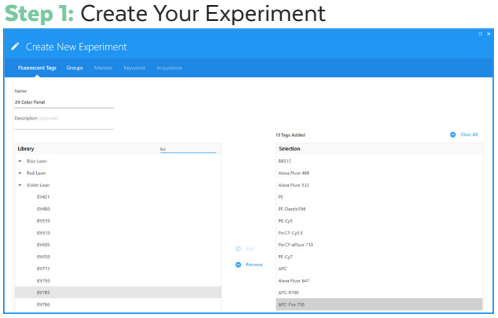
Users:
For administrative controls.

Preferences:
Customize the software appearance. Set default plot sizes, text sizes and fonts, gate colors, print layout, statistics table options, and more.

Experiment Workflow:

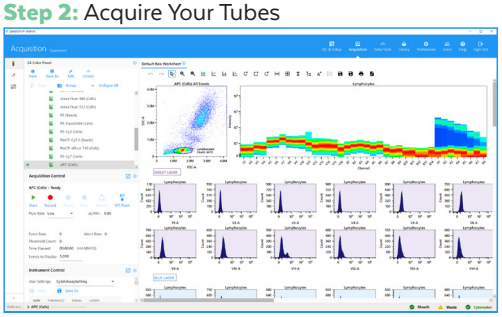
From the Acquisition menu, you can start a new experiment and get to your data in four simple guided steps.

Step 1: Create Your Experiment



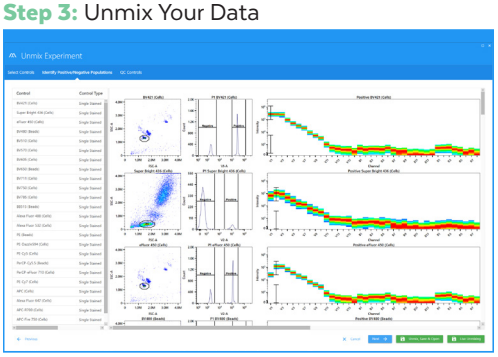
Create your experiment, choose fluorochromes, and add labels, tubes, worksheets, and stopping criteria in this guided workflow.

Step 2: Acquire Your Tubes



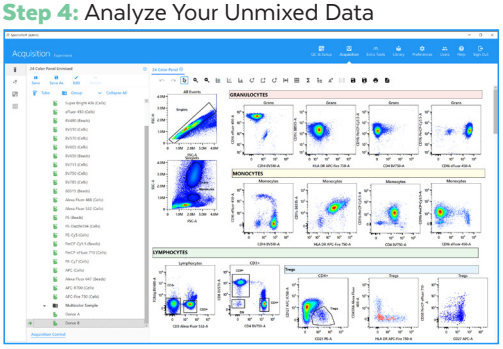
Load and run your tubes.

Step 3: Unmix Your Data



Visualize your reference control spectra with the unmixing wizard.

Step 4: Analyze Your Unmixed Data



Create an analysis worksheet and save it as a template to reuse and share with others.

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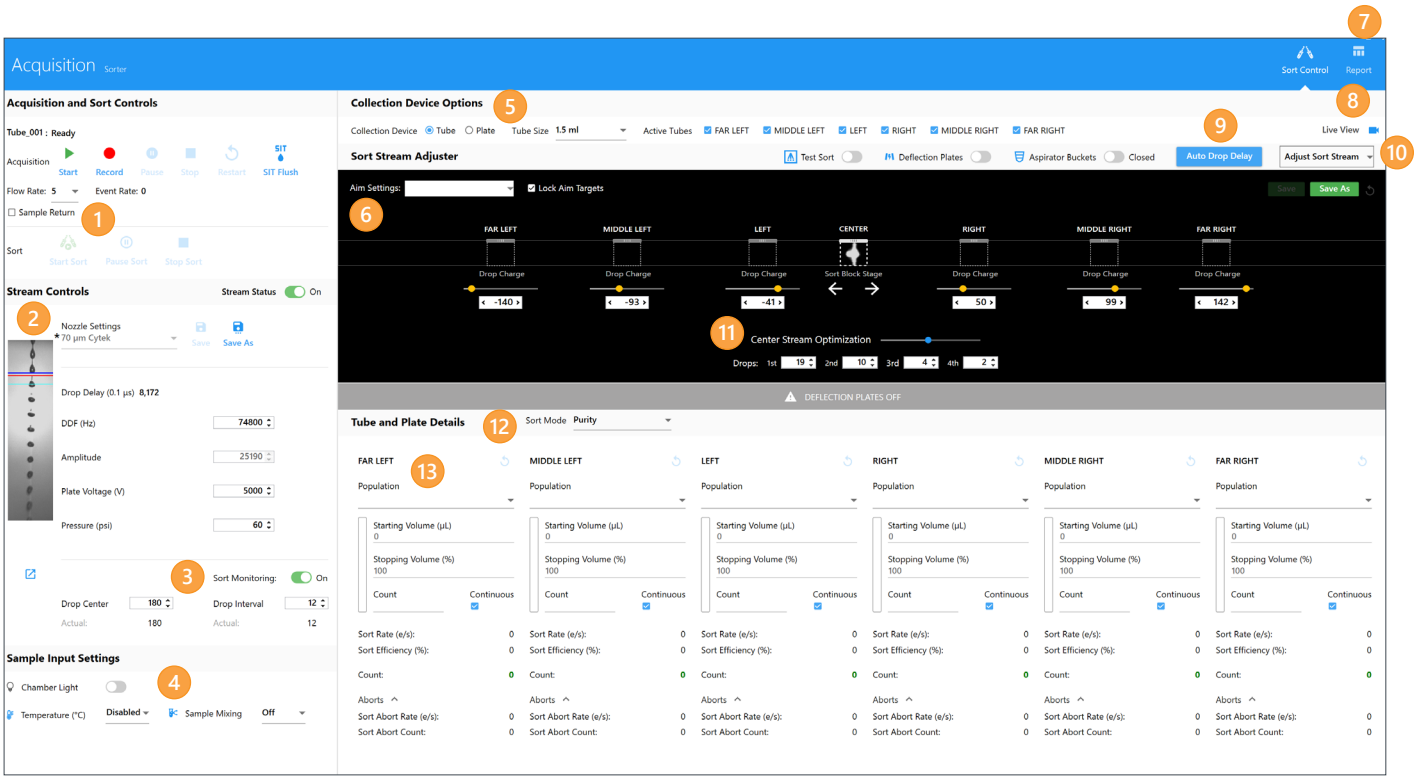
Sort Controls Inside SpectroFlo® CS Software

The Sorter Control window inside SpectroFlo® CS software has a variety of controls and guided workflows highlighted below to help each user achieve a successful sort output.



1. Enable sample return.
2. Select nozzle size, optimize drop break-off and stream settings, and save settings for future use.
3. Enable sort monitoring with clog detection to help ensure a successful sort.
4. Enable sample mixing and temperature control.
5. Select tube or plate sort collection device.
6. Adjust tube and plate aiming targets and save aim settings for future use.
7. Post-sorting, view and export a comprehensive sort report.
8. Use the Live View camera feed to help aim streams into the center of each tube and monitor sort tube volumes.
9. Click the Auto Drop Delay button to open the automated drop delay wizard.
10. Toggle to Adjust Sort Stream or Adjust Drop Delay manually.
11. Use the slider and/or manually change values in the text boxes to optimize the center stream.
12. Choose a Cytek-defined sort mode or create a custom mode.
13. Define population(s) and sort stopping criteria for each plate well or tube.

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Technical Specifications

Optics

EXCITATION OPTICS

OPTICAL PLATFORM
Aurora CS system contains a fixed optical assembly with the capacity to be configured with up to five spatially separated laser beams. Laser delays are automatically adjusted during instrument QC.

LASERS
Base model three-laser configuration: 405 nm: 100 mW, 488 nm: 50 mW, 640 nm: 80 mW
Available laser upgrades: 355 nm: 20 mW, 561 nm: 50 mW

BEAM GEOMETRY
Flat-Top laser beam profile with narrow vertical beam height optimized for small particle detection.

EMISSION OPTICS

EMISSION COLLECTION
Fused silica cuvette coupled to high NA lens for optimum collection efficiency to optical fibers.

FORWARD AND SIDE SCATTER DETECTION
FSC: high-performance semiconductor detector with 488 nm bandpass filter
SSC: two high-performance semiconductor detectors with 405 nm and 488 nm bandpass filters

FLUORESCENCE DETECTORS
Proprietary high sensitivity Coarse Wavelength Division Multiplexing (CWDM) semiconductor array per laser enabling more efficient spectrum capture in the 365-829 nm range. No filter changes required for any fluorochrome excited by the 355 nm, 405 nm, 488 nm, 561 nm, 640 nm lasers.

STANDARD OPTICAL CONFIGURATION
Violet detector module: 16 channels unevenly spaced bandwidth from 420-829 nm.
Blue detector module: 14 channels unevenly spaced bandwidth from 498-829 nm.
Red detector module: 8 channels unevenly spaced bandwidth from 652-829 nm.

4 and 5 Laser Options:
Yellow-Green detector module: 10 channels unevenly spaced bandwidth from 567-829 nm.
Ultraviolet detector module: 16 channels unevenly spaced bandwidth from 365-829 nm.

Fluidics

SAMPLE FLOW RATES
Adjustable in increments of 7 µL/min from 10 µL/min to 80 µL/min

FLUIDIC MODES
Fluidics startup, Fluidics shutdown, SIT flush, Purge filter, Clean flow cell, Long clean, Sample line return

SAMPLE INPUT FORMATS
12x75 mm or 15 mL polystyrene or polypropylene tube with sample mixing

FLUIDIC RESERVOIRS
10 L sheath and waste fluid containers with level-sensing provided

Electronics

SIGNAL PROCESSING
Digital signal processing with automatic window gate adjustment.

22-bit 6.5 log decades.

Threshold using any single parameter or combination of parameters.

PULSE SHAPE PARAMETERS
Pulse Area and Height for every parameter. Width for scatter parameters and one fluorescence parameter for each laser.

Workstation

Workstation specifications may vary between laser configuration; below is for three-laser configuration.

OPERATING SYSTEM
Windows® 10 Pro 64-bit

PROCESSOR
Intel® Core™ i7 processor, 3.6 GHz

RAM
64 GB

HARD DRIVE
500GB SSD and 1 TB SATA

MONITOR
Two 24" UHD 4K Monitors

Preliminary Performance*

*Formal verification pending.

FLUORESCENCE LINEARITY
FITC R^2 ≥0.995 / PE R^2 ≥0.995

FORWARD AND SIDE SCATTER RESOLUTION
Performance is optimized for resolving lymphocytes, monocytes, and granulocytes.

SIDE SCATTER RESOLUTION
Capable of resolving 0.1 µm polystyrene beads from noise.

CARRYOVER
≤0.1%

DATA ACQUISITION RATE
25,000 events/s**
**Five-laser system

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Functional Specifications

Sort Output

SORT COLLECTION

Up to 4-way sorting: 5 mL, and 1.5 mL polystyrene and polypropylene tubes.

Up to 6-way sorting: 1.5 mL polystyrene and polypropylene tubes.

96-well plates with index sorting.

NOZZLES

Quick-replace 70, 100, and 130 µm* nozzles with optimized and user definable pressure and sorter settings.

*130 µm available on request

SORT MODES

Multiple optimized sort modes for purity, enrichment, mixed, and single cell plus user definable sort modes.

Deposit 1 cell per well into 96 wells in less than 2 minutes.

Temperature Control

4 to 30˚C for both sample input and output.

Biosafety

PRIMARY

Built-in aerosol management with user replaceable HEPA filters.

SECONDARY

Optional Class II, Type A2 Biosafety Cabinet specifically designed for Cytek Aurora Sorter and tested to major worldwide Biosafety Standards with sorter inside.*

* Not manufactured by Cytek

Software

SPECTROFLO® CS SOFTWARE

Live unmixing during acquisition

Sort on raw or unmixed data

Developed specifically to streamline assay setup, data acquisition, and file export

Automated QC module

Autofluorescence extraction

Automated drop delay module

Default and customizable sort modes and nozzle settings

Sort collection tube volume monitoring and live view

Autogenerated sort reports

Raw and Unmixed FCS 3.1 files

Regulatory

Class 1 Laser Product.
For Research Use Only. Not for use in diagnostic or therapeutic procedures.

Installation Requirements

Dimensions (W x D X H)

INSTRUMENT DIMENSIONS

75 x 57 x 65 cm

INSTRUMENT WEIGHT

105 kg

BIOSAFETY CABINET DIMENSIONS

150 x 91 x 231 cm

RECOMMENDED WORKSPACE

183 x 81 x 94 cm

Room Requirements

POWER

100-140 VAC, 15A or 200-250 VAC, 10A

HEAT DISSIPATION

1000 W with all solid-state lasers

TEMPERATURE

Outside of Biosafety Cabinet: 18-28˚C

Inside Biosafety Cabinet: 18-26˚C

HUMIDITY

20%-85% relative non-condensing

AIR SUPPLY

551.5 to 586 kPa (80 to 85 PSI) clean dry air

AIR FILTERING

No excessive dust or smoke

LIGHTING

No special requirements

Preliminary Sort Performance*

*Formal verification pending.

SORT PURITY

1%-2% population of lymphocytes using a 70 µm nozzle, mixed sort mode, and a system threshold rate of 20,000 events/second:

Sort purity ≥ 95% and sort yield is ≥ 90% to theoretical yield.

SORT GATES

Sort up to 6 populations up to 64 levels deep in the gating hierarchy.

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Cytek Biosciences is dedicated to enhancing our customers' user experience. The Cytek Aurora CS system is backed by our world-class service and support team that can provide phone or field-based assistance. Various levels of maintenance options are available to meet your needs now, and in the future.

For more information: email us at sales@cytekbio.com or call 1-877-922-9835

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