Advanced Flow Cytometry and Cell Sorting Training Course

Professional Development Training Course Outline

Category	Biotechnology and Pharmaceutical Development	**Duration**	10 Days
Base Fee	$4,000 USD	**Delivery Mode**	Online / On-site Hybrid

Course Introduction

Advanced Flow Cytometry and Cell Sorting Training Course moves beyond the fundamentals to focus on mastering high-dimensional data acquisition and Fluorescence-Activated Cell Sorting, addressing the critical demand for specialized expertise in modern life sciences. The curriculum emphasizes the newest technologies, including Spectral Flow Cytometry and High-Parameter Panel Design, to tackle complex biological questions in immunology, oncology, and regenerative medicine. Participants will gain hands-on proficiency in advanced instrument troubleshooting, complex compensation/unmixing, and the execution of delicate single-cell sorting protocols for subsequent 'Omics' applications. This training is essential for researchers and lab personnel aiming to lead experiments involving rare event detection, complex immunophenotyping, and isolating viable cell subsets for clinical or translational research.

This program uniquely bridges the gap between sophisticated instrument operation and Flow Cytometry Bioinformatics. A major focus will be placed on unsupervised analysis techniques like t-SNE, UMAP, and FlowSOM, enabling unbiased discovery of novel cell populations in high-parameter datasets, such as those derived from the tumor microenvironment or CAR-T cell therapy products. By integrating best practices for Quality Control (QC), standardization, and GLP/GCP compliance, attendees will learn to generate reproducible, publication-quality data. Successful completion will elevate participants to independent, expert-level users capable of designing and executing cutting-edge flow cytometry and cell sorting experiments that directly accelerate biomarker discovery and therapeutic development.

Programme Curriculum

Advanced Flow Cytometry and Cell Sorting Training Course

Introduction

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Course Objectives

1. Master the principles and practical application of Spectral Flow Cytometry and Full-Spectrum Analysis.
2. Design and rigorously optimize High-Parameter Panels using Spillover Spreading Matrix metrics.
3. Perform accurate Spectral Unmixing and troubleshoot complex compensation/unmixing errors.
4. Develop Advanced Gating Strategies for accurate Rare Event Detection and minimal false positives.
5. Execute Fluorescence-Activated Cell Sorting protocols for high-purity, high-viability Single-Cell Isolation.
6. Apply High-Dimensional Data Analysis methods.
7. Design and analyze advanced functional assays.
8. Implement robust Quality Control and standardization protocols for instrument calibration and assay consistency.
9. Troubleshoot complex fluidic, optical, and electronic issues unique to high-speed Cell Sorters.
10. Integrate flow cytometry data with other Single-Cell 'Omics' techniques.
11. Interpret and effectively communicate complex multivariate flow cytometry datasets for Publication.
12. Explore specialized applications in CAR-T Cell manufacturing and Regenerative Medicine.
13. Utilize Automated Gating and Machine Learning algorithms for objective cell population identification.

Target Audience

1. Senior Research Scientists and Postdoctoral Fellows.
2. Core Facility Managers and Lead Technicians.
3. R&D Scientists in Biotechnology and Pharmaceutical companies.
4. Clinical Trial Investigators and Technicians.
5. Graduate Students with foundational flow cytometry experience.
6. Experienced researchers migrating from conventional to Spectral Flow Cytometers.
7. Laboratory personnel performing High-Purity Cell Sorting for downstream Genomic/Proteomic analysis.

8. Immunologists and Oncologists.

Course Modules

Module 1: Advanced Principles of Light, Optics, and Fluidics

- Principles of Spectral Flow Cytometry.
- Understanding and optimizing Laser configuration, alignment, and power settings.
- In-depth look at Fluidics and its impact on sort purity/viability.
- Impact of sample concentration and acquisition rate on data quality
- Case Study: Evaluating the trade-offs of using a high-pressure sorter for fragile stem cells versus a jet-in-air system.

Module 2: High-Parameter Panel Design and Optimization

- Strategy for fluorochrome selection using the Spillover Spreading Matrix
- Rules for pairing weak vs. bright antigens with appropriate fluorochromes.
- Importance and execution of proper Antibody Titration for every reagent.
- Designing comprehensive panels for complex Immunophenotyping
- Case Study: Designing a 28-color panel for simultaneous B-cell and T-cell subset analysis, focusing on minimal spectral spread.

Module 3: Advanced Compensation and Spectral Unmixing

- Understanding the root cause of Spillover and Spread.
- Troubleshooting over- and under-compensated data in both conventional and spectral systems.
- The math and theory behind Spectral Unmixing and its critical assumptions.
- Best practices for creating and validating robust single-stained controls (SSCs) for unmixing.
- Case Study: Correcting unmixing artifacts in a complex panel containing multiple tandem dyes and autofluorescence.

Module 4: Quality Control (QC) and Standardization

- Tracking laser delay, sensitivity, and CVs using standardized beads.
- Establishing and tracking a Cytometer Setup and Tracking baseline for standardization across experiments.
- Importance of FCS file standards and metadata annotation.
- Implementation of GCP/GLP-compliant documentation and SOPs for clinical studies.
- Case Study: Using Levey-Jennings plots and standardization beads to track instrument stability over a multi-week clinical trial.

Module 5: Principles of Fluorescence-Activated Cell Sorting (FACS)

- Physics of droplet formation, break-off, and the role of the Piezo Crystal.
- Detailed understanding of the Electrostatic Deflection process and charging delay.
- Drop-Delay calibration, amplitude, and purity/yield mode selection.
- Safety considerations: Aerosol containment and biohazard protocols for cell sorting.
- Case Study: Optimizing sort parameters to achieve purity while maintaining viability for a downstream cell culture experiment.

Module 6: High-Purity Single-Cell Isolation

- Techniques for sorting extremely Rare Cell Populations
- Optimizing for high viability for post-sort functional assays and cell culture.
- Setting up the sorter for Single-Cell Sorting into 96-well or 384-well plates.
- Verification of single-cell deposition and minimization of doublets/triplets.
- Case Study: Isolating single B-cells into PCR strips for subsequent scRNA-Seq analysis and clonal lineage tracing.

Module 7: Advanced Sample Preparation and Controls

- Protocols for tissue dissociation to ensure high single-cell suspension quality.
- Staining protocols for Intracellular Staining (ICS) and Phospho-Flow.
- Critical use of Fluorescence Minus One (FMO) and Isotype controls for complex gating.
- Managing cell clumping, debris, and non-specific staining in challenging samples.
- Case Study: Developing a sample preparation protocol for a solid tumor biopsy to maximize cell yield and minimize autofluorescence.

Module 8: Advanced Data Management and FCS Files

- Understanding the structure of FCS files, including the TEXT, DATA, and ANALYSIS segments.
- Best practices for data backup, archiving, and cloud-based storage.
- Setting up an organized data repository and using batch processing for multi-experiment data.
- Strategies for data anonymity and HIPAA/GDPR compliance in clinical studies.
- Case Study: Implementing a standardized file naming and keyword annotation system across an entire research team for easier data retrieval.

Module 9: High-Dimensional Data Visualization: t-SNE and UMAP

- Theoretical basis for Dimensionality Reduction algorithms like t-SNE and UMAP.
- Choosing appropriate parameters and optimizing the 'Perplexity' and 'Neighbors' settings.
- Interpreting the clustered maps and identifying potential technical/biological artifacts.
- Using Heatmaps and other visualization tools to map marker expression onto clusters.
- Case Study: Visualizing a 15-parameter mouse splenocyte dataset to identify and annotate rare dendritic cell subsets using a UMAP plot.

Module 10: Unsupervised Clustering and Automated Gating

- Introduction to clustering algorithms.
- Workflow for Unsupervised Analysis to enable unbiased cell population discovery.
- Validating computationally derived clusters with biological knowledge and controls.
- Transitioning from manual gating to objective, Automated Gating strategies.
- Case Study: Applying FlowSOM to a complex immune dataset from an autoimmune disease model to discover novel, disease-specific cell clusters.

Module 11: Functional Assays: Intracellular and Phospho-Flow

- Detailed fixation and permeabilization protocols for successful Intracellular Staining (ICS).
- Designing panels for simultaneous surface, cytokine, and transcription factor analysis.
- Protocols for monitoring intracellular signaling pathways using Phospho-Flow Cytometry.
- Kinetic analysis: Measuring calcium flux and receptor internalization dynamics.
- Case Study: Analyzing the phosphorylation of STAT proteins in T-cells following a specific cytokine stimulation time course.

Module 12: Applications in Immuno-Oncology and Cell Therapy

- Detailed Immunophenotyping of Tumor-Infiltrating Lymphocytes (TILs) and their exhaustion status.
- Quality control and potency assessment of CAR-T Cell and NK Cell products.
- Monitoring residual disease and minimal residual disease (MRD) in hematological malignancies.
- Assays for assessing cytotoxicity and target cell killing efficiency.
- Case Study: Designing a potency assay for a clinical-grade CAR-T cell product based on T-cell activation and proliferation markers.

Module 13: Applications in Stem Cells and Regenerative Medicine

- Identification and sorting of Hematopoietic Stem Cells (HSCs) and Mesenchymal Stem Cells (MSCs).
- Protocols for viability assessment and functional analysis of sorted stem cells.
- Characterization of pluripotency and differentiation markers in iPSCs.
- Advanced sorting for organoid generation and therapeutic delivery.
- Case Study: Isolating CD34+ stem cells from mobilized peripheral blood for autologous transplantation, focusing on high purity and yield.

Module 14: Integrating Flow Cytometry with Omics Technologies

- Sample preparation considerations for linking flow sort to scRNA-Seq
- Utilizing Antibody-Oligonucleotide Conjugates for protein/RNA correlation.
- Data integration strategies.
- Practical considerations for sorting low-input samples for Proteomic analysis.
- Case Study: Designing a CITE-seq experiment to simultaneously measure 10 surface proteins and the full transcriptome of sorted immune cells.

Module 15: Troubleshooting and Data Reporting for Publication

- Systematic approach to troubleshooting common issues
- Best practices for statistical analysis and hypothesis testing on flow data.
- Creating publication-quality figures, including appropriate axis scaling
- Checklist for reporting flow cytometry experiments according to MIFlowCyt standards.
- Case Study: Reviewing a complete flow cytometry experiment and identifying 5 critical errors that would compromise publication.

Training Methodology

The course utilizes a blended, immersive learning approach:

- Interactive Lectures
- Hands-on Instrument Sessions.
- Data Analysis Workshops
- Real-World Case Studies
- Troubleshooting Labs.

Register as a group from 3 participants for a Discount

Send us an email: info@fineskilltrainingcenter.org or call +254769199797

Certification

Upon successful completion of this training, participants will be issued with a globally- recognized certificate.

Tailor-Made Course

We also offer tailor-made courses based on your needs.

Key Notes

a. The participant must be conversant with English.

b. Upon completion of training the participant will be issued with an Authorized Training Certificate

c. Course duration is flexible and the contents can be modified to fit any number of days.

d. The course fee includes facilitation training materials, 2 coffee breaks, buffet lunch and A Certificate upon successful completion of Training.

e. One-year post-training support Consultation and Coaching provided after the course.

f. Payment should be done at least a week before commence of the training, to Fineskill Training Center account, as indicated in the invoice so as to enable us prepare better for you.

Upcoming Scheduled Sessions

Start Date	End Date	Location	Price
23 Nov 2026	04 Dec 2026	Online	$2,000
23 Nov 2026	04 Dec 2026	Physical	$3,000
23 Nov 2026	04 Dec 2026	Physical	$6,000
23 Nov 2026	04 Dec 2026	Physical	$6,000
23 Nov 2026	04 Dec 2026	Physical	$12,000
23 Nov 2026	04 Dec 2026	Physical	$9,000
23 Nov 2026	04 Dec 2026	Physical	$12,000
23 Nov 2026	04 Dec 2026	Physical	$12,000

Ready to enroll or request a customized program? Book online or contact us:

Register Online Now