



Think outside the dot.

Insight & Analysis

Welcome to a Revolution in Cell Analysis: Imaging Flow Cytometry

The ImageStream® system advances your science by combining quantitative cellular imagery with powerful population statistics.

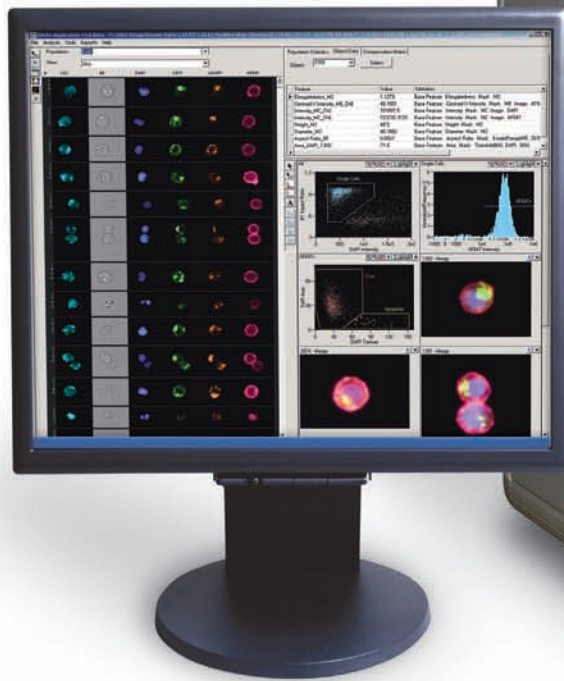
With traditional cell analysis, you've had to choose between visualizing a few cells under a microscope without quantitation or analyzing large cell populations by flow cytometry without imagery. Microscopy gives you detailed fine structure, morphology, and qualitative molecular localization. Flow cytometry gives you robust statistical information and detection of rare sub-populations. No technology has been able to give you all this at the same time and in a single experiment.

Until now.

The ImageStream system from Amnis gives you morphology, fluorescence localization, *and* population statistics for a broad range of applications. Quantitate translocation of transcription factors between cellular compartments. Examine interactions in cell conjugates. Perform high throughput FISH. Examine cells in mitosis and apoptosis. Quantitate internalization. Study the distribution and abundance of fluorescent proteins.

And do it all in rare cells and highly heterogeneous samples.

amis
ImageStream



New Technology

An Entirely New Way to Analyze Cells

It's all about the numbers: 500 quantitative parameters from six simultaneous images per cell; brightfield, darkfield and multiple fluorescence images; 0.75 NA optics; sub-micron resolution; over 15,000 cells per minute.

ADVANCED DETECTION TECHNOLOGY DELIVERS HIGH SENSITIVITY WITH HIGH SPEED

A unique six-channel CCD camera and a novel velocity detection system work in concert to collect 1,000 times more light than conventional technology. The technique is called Time Delay Integration. The result is high resolution imagery with fluorescence sensitivity superior to flow cytometry.

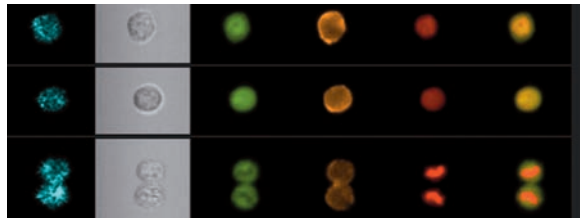
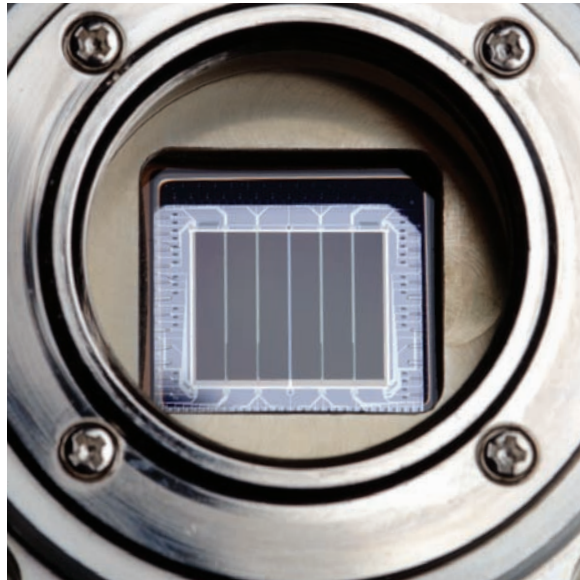
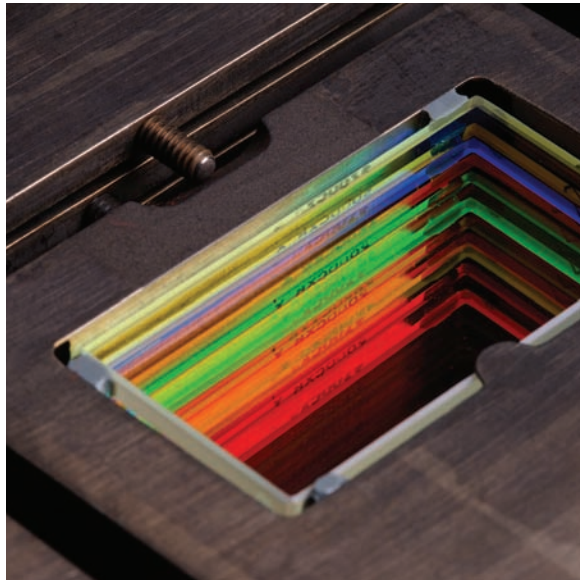
MULTISPECTRAL IMAGING FOR MAXIMUM INFORMATION PER CELL

Illumination in the ImageStream system is provided by a brightfield lamp, a 488 nm laser and optional violet and red excitation lasers. Cellular imagery is split into six component

colors through a unique spectral decomposition element. The result is a brightfield image, a darkfield image, and multiple fluorescence images of every cell.

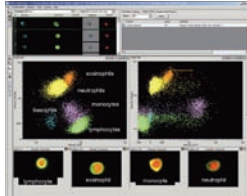
SIMPLE OPERATION

ImageStream technology is sophisticated, but operating the instrument isn't difficult. Highly automated protocols step you through calibration and set up. Focus and cell tracking are automatic. A single click of the mouse sterilizes the instrument. We've designed the ImageStream to allow you to concentrate on your research, not your instrumentation.

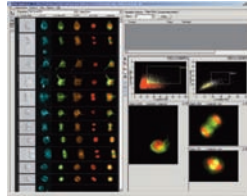


New Applications

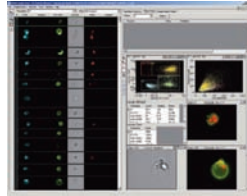
Leukocyte cell classification



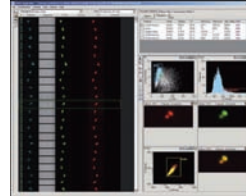
Tubulin stain in adherent cells



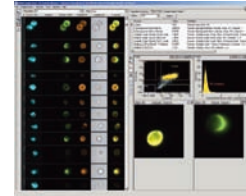
Apoptosis



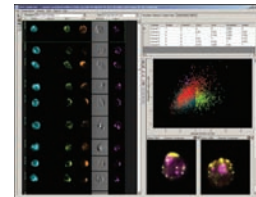
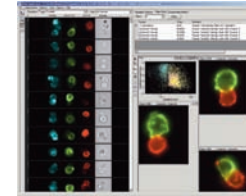
Yeast budding



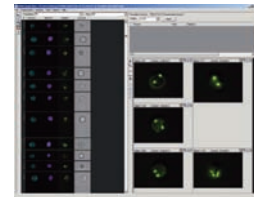
Surface marker capping



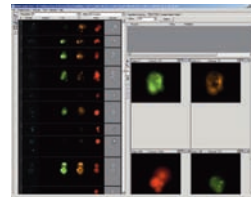
T Cell / APC conjugates



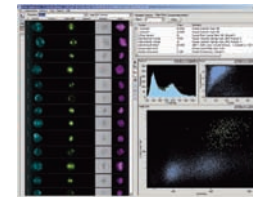
Endocytic processing pathways



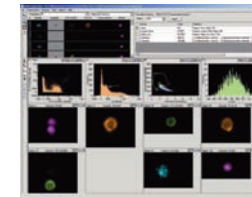
Multicolor FISH in Suspension



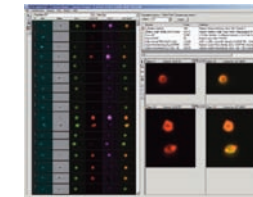
GFP and apoptosis



Cell cycle analysis



Human PBMC



Immune synapse

One Instrument, Many Applications

VERSATILE

AMNIS

CELL SIGNALING / PATHWAY ANALYSIS

The ImageStream system brings significant new capabilities to pathway analysis for cells in suspension. The phosphorylation states of key signaling molecules and their locations within the cell can be measured directly. Molecular association with the cell membrane, the cytoplasm, or the nucleus is easily distinguished and quantitated.

ANALYSIS OF CELL CONJUGATES

Cells communicate through cell membrane-mediated molecular interactions. The ImageStream system not only identifies cell doublets, but also quantitates molecular co-localization at the interface between the interacting cells.

FLUORESCENCE IN SITU HYBRIDIZATION IN SUSPENSION (FISHIS)[®]

High throughput FISH is now possible with cells in suspension using the ImageStream system and Amnis protocols. Imagery is acquired rapidly and extended depth of field technology provides exceptionally clear visualization of multicolor chromosome spots in a range of cell types.

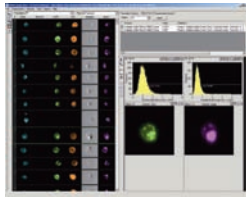
INTERNALIZATION AND INTRACELLULAR TRAFFICKING

Image similarity algorithms allow you to quantitatively compare the distribution of multiple signals within single cells for co-localization, co-capping, and similar studies.

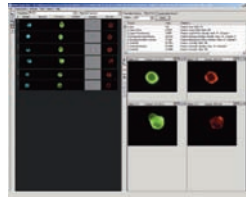
GENE EXPRESSION ANALYSIS

The ImageStream system is well suited to the analysis of Green Fluorescent Protein and other fluorescent markers used in the study of gene expression. The high spatial resolution and sensitivity of the ImageStream system allows quantitation of expression levels and localization of expression to specific regions of the cell and key organelles.

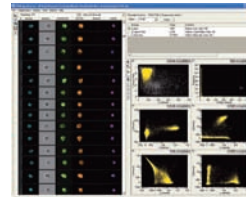
Intracellular co-localization of multiple proteins



HIV receptor mapping



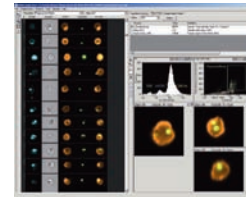
Multiple surface receptors



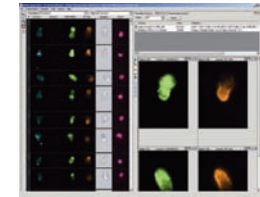
Nf-κB translocation to the nucleus



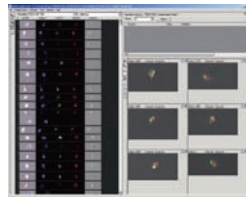
Phagocytosis



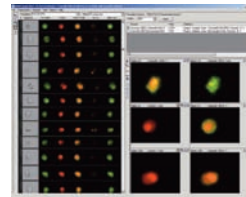
Pseudopod formation



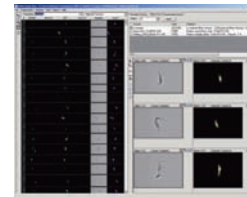
Nuclear translocation of Raf



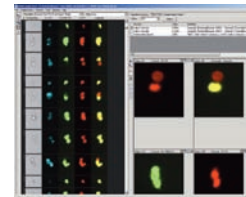
FISH in sperm



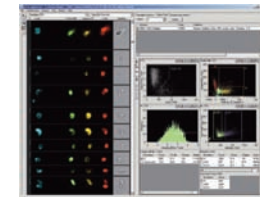
Toxoplasma gondii



Gene expression in trypanosomes



Analysis of TUNEL+ and TUNEL- cells



Caspase and NF-κB in apoptosis

RECEPTOR MAPPING AND DISTRIBUTION

The ImageStream system not only measures the abundance of important cell surface receptors with exceptional sensitivity and resolution, but can also map their locations and co-localize them with ligands or intracellular organelles. For instance, proteins of interest may be co-localized with endosomal and lysosomal markers to follow intracellular processing and degradation.

QUANTITATIVE MORPHOLOGY

Change in cell shape is closely correlated with function in the analysis of lymphocyte or macrophage activation, pseudopod formation, response to drugs, and many other instances. Powerful features in the IDEAS® image analysis software allow you to accurately classify cells based on shape and structure.

CELL CLASSIFICATION

Characterization of peripheral blood mononuclear cell populations is a fundamental tool in hematology. The ImageStream system combines classical surface phenotyping with morphologic classification to deliver a full five-part differential analysis with room for the identification of additional sub-populations using fluorescent markers.

APOPTOSIS

Using only measurements of nuclear morphology, the ImageStream system can directly differentiate apoptotic and necrotic cells, quantify the extent of apoptosis in cell populations, and calculate sub-population frequencies. The need for surrogate markers such as Annexin V or fluorescent caspase substrates is reduced or eliminated, as are classification errors found in conventional flow cytometric apoptosis assays, reducing false positive and false negative results.

Analytical Power

Designed by Biologists for Biologists

Powerful, flexible, and extremely easy to learn, the IDEAS statistical image analysis package is integral to the ImageStream system.

A ROBUST FEATURE SET, EXPANDABLE TO MEET YOUR NEEDS

The IDEAS feature set – the heart of the image analysis package – is extraordinarily robust, providing more than 500 features for every cell. IDEAS also allows you to create almost any new feature you find useful (e.g. nuclear to cytoplasmic area).

IMAGE DATA AND STATISTICAL DATA ARE FULLY INTEGRATED

In IDEAS, graphs and imagery are completely integrated. Every dot on a scatter plot links directly to a cell's images – click on the dot and you'll see the corresponding cell. With its virtual sorting capability, IDEAS will show you all the images of a cell population you define.

AN EFFICIENT, FLEXIBLE DATA INTERFACE

The IDEAS interface integrates image data, plots, and statistics. The Gallery shows you images of every cell, while the Workspace gives you graphing tools to define and analyze cell populations. The Tabular Data section allows you to view population statistics as well as individual feature values.

TEMPLATES AND BATCH PROCESSING

Once you've created an analysis scheme in IDEAS, you can save it as a template for batch processing future experiments or to share with your colleagues.



SIMPLE, FLEXIBLE POPULATION DEFINITIONS

Easy to use gating tools allow you to define, name and visualize cell populations quickly and intuitively.

RICH FEATURE SET

IDEAS calculates over 80 features for each image and over 500 features per cell, allowing the discrimination of subtle differences between cell populations.



FAMILIAR GRAPHING TOOLS

Quickly and easily create scatter plots and histograms to define your cell populations. Display parameters are easily adjusted.

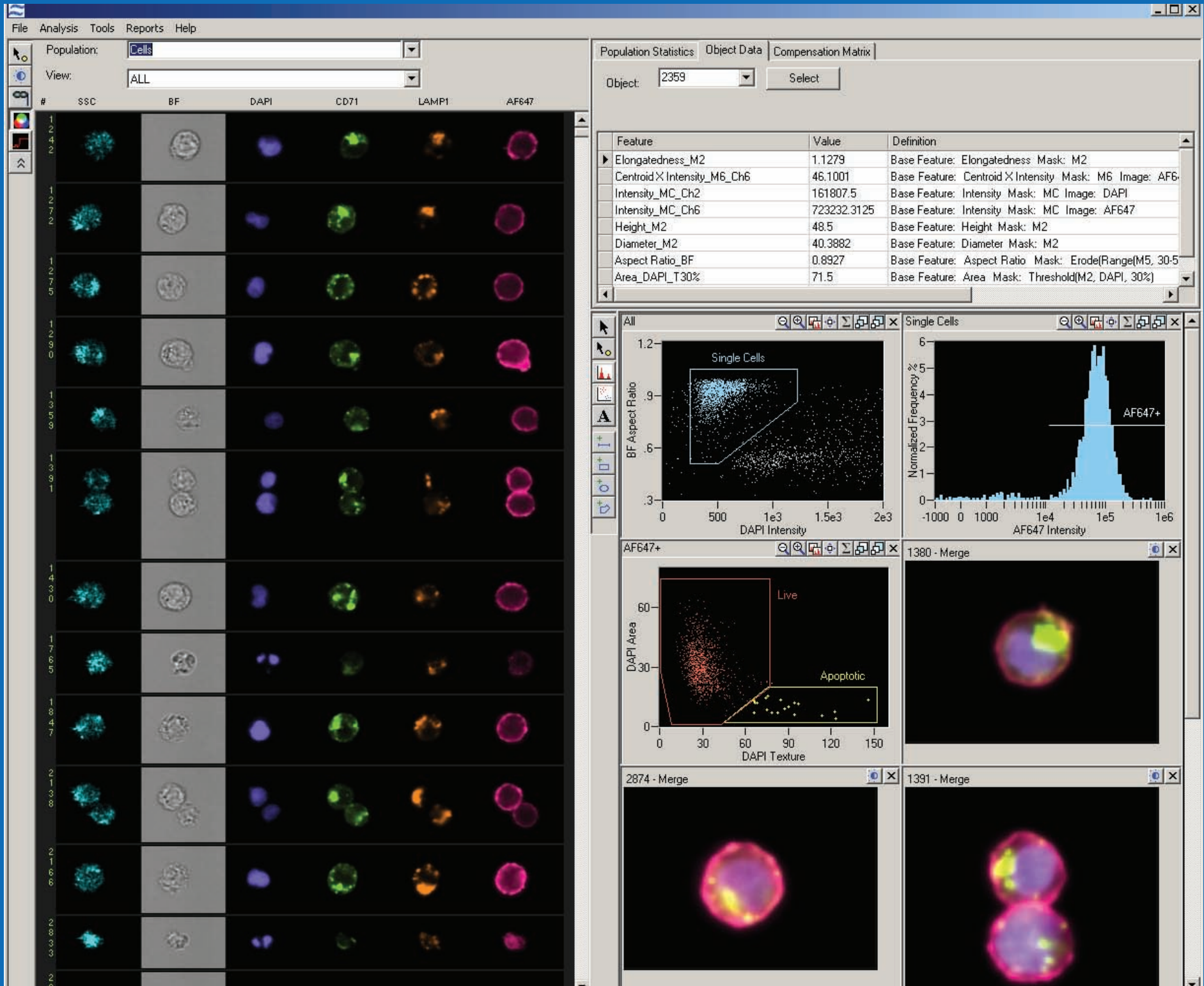


DATA LINKAGE

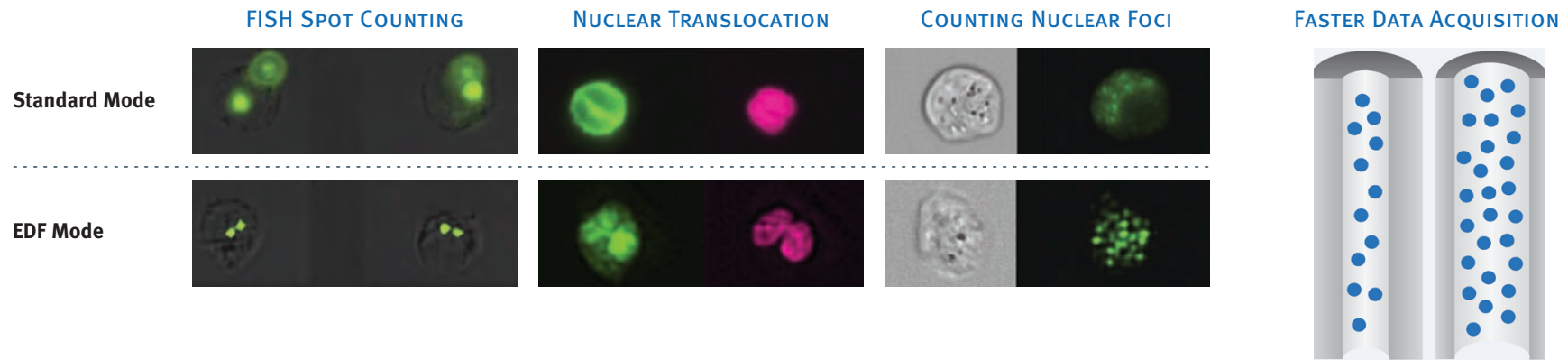
Every dot in a scatter plot is linked to a set of cell images. Click on the dot to see the cell, or click on a cell's images to locate it in all plots.

QUANTITATIVE DATA PLOTTING

Any image analysis feature can be used in a histogram or dot plot. Extend your analysis beyond simple fluorescence intensity with localization and morphology features.



Advanced Performance



EDF™ technology breaks the classical depth of field barrier

IMAGE THE WHOLE CELL IN FOCUS

EDF extended depth of field technology uses a combination of specialized optics and unique image processing algorithms to project all structures within the cell into one crisp plane of focus.

ENABLES NEW APPLICATIONS

Many applications, such as FISH, depend critically on the resolution and accurate counting of spots within the cell. With the exceptional focus depth of EDF, high throughput analysis of FISH has become a reality.

IMPROVED PRECISION AND DISCRIMINATION

In addition to increasing depth of field, EDF improves resolution, thereby enhancing the discrimination of cellular features and improving precision in the quantitative analysis of cell imagery over a wide array of applications.

REDUCED DATA ACQUISITION TIME

In addition to keeping the whole cell in focus, the EDF option allows the ImageStream to be run with a larger core diameter, thereby increasing throughput by up to three-fold.

THE EDF EXTENDED DEPTH OF FIELD OPTION

The EDF option for the ImageStream system includes all required modifications to the instrument and software, installation, testing, documentation and user training. The EDF option can be included with a new ImageStream system or installed as a field upgrade.

ImageStream Specifications

Advanced Engineering Creates Exceptional Performance

PERFORMANCE

- Imaging rate: up to 300 cells/second
- Sample throughput: ~5 min/sample
- Detection limit: <50 fluorescent molecules
- Numeric aperture: 0.75
- Pixel size: 0.5 x 0.5 microns
- Field of view: 45 microns wide
- Sample volume: 40-200 microliters

DATA ANALYSIS

- Automated crosstalk compensation post-acquisition
- Unlimited user-defined image features
- Over 500 standard image features per cell

INSTRUMENT OPERATION

- Automated sample load, empty, flush, and purge
- Automated focus and core position tracking
- Automated sterilization
- Automated calibration and quality control
- Automated laser alignment

REQUIREMENTS

- 90-240 VAC, 50-60 Hz
- 100 Mbps ethernet, minimum
- No external air or water required
- 36" w x 24" h x 24" d
- 350 lbs

ILLUMINATION SOURCES

SOURCE		WAVELENGTH	MAX POWER
Brightfield Lamp	Standard	430-730 nm	
Blue Laser	Standard	488 nm	200 mW
Violet Laser	Optional	405 nm	350 mW
Red Laser	Optional	658 nm	80 mW

The ImageStream system includes a solid-state 488 nm laser (200 mW) as the standard excitation source. The laser options include a red 658 nm laser (80 mW) and a high power violet 405 nm laser (350 mW). Each of these laser options is available factory installed or they may be purchased for installation on an existing ImageStream system.

The dyes listed here represent just some of those that may be used on the ImageStream system configured with 405 nm, 488 nm and 658 nm lasers. The ImageStream is a Class 1 laser product.

DETECTION CHANNELS

CHANNEL 1 488 nm	CHANNEL 2 430-470 nm	CHANNEL 3 500-560 nm	CHANNEL 4 560-595 nm	CHANNEL 5 595-660 nm	CHANNEL 6 660-735 nm
Darkfield	DAPI	FITC	PE	7-AAD	Cy5, Cy5.5
	Hoechst 33258	Alexa Fluor 488	Cy3	PE-Alexa Fluor 610	CyChrome
	Hoechst 33342	Alexa Fluor 500	Alexa Fluor 546	Propidium Iodide	Alexa Fluor 647
	Alexa Fluor 405	Alexa Fluor 514	Alexa Fluor 555	PE-Texas Red	Alexa Fluor 660
	Alexa Fluor 430	Syto 11, 13, 16	YFP	Qdot 605	Alexa Fluor 680
	Cascade Blue	Mitotracker Green	OFP	Qdot 625	Alexa Fluor 700
	Pacific Blue	Spectrum Green	Qdot 565	ECD	DRAQ5
	LIVE/DEAD Violet	Lucifer Yellow	Qdot 585	Brightfield	PerCP
	Vybrant DyeCycle Blue	Cascade Yellow	POPO-3		Qdot 705
	CFP	Qdot 525	PO-PRO-3		APC
	Brightfield	Qdot 545	DsRED		Mitotracker Deep Red
		Brightfield	Brightfield		Brightfield

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