Time-Resolved Flow Cytometry: Expanding the Power of Cell Analysis
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Flow Cytometry: Cell-by-Cell Interrogation

- Laser(s)
- Excitation light
- Flowcell
- Sheath
- Cells
- Fluorescence
- Detectors
Flow Cytometry: At a Glance

**Technical**
- fluorescence and light scattering
- 2–10 lasers (UV–visible)
- 4–30+ detectors: PMTs, PDs, APDs, arrays
- typ. 4–12 markers/cell (but up to 30+)
- ~ 10–30k cells/second

**Market**
- $5.3B/yr
- growing at 10%/yr
- hematology: add’l $4B/yr, growing at 4%/yr

(Some) Flow Cytometry Applications

- drug discovery
- immunophenotyping
- cell proliferation
- organ transplant
- milk testing
- exosomes
- HIV testing
- malaria testing

- oncology
- hematology
- stem cells
- microbiology
- milk testing
- exosomes
- HIV testing
- malaria testing

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Cells Are Labeled With Fluorophores

excitation light

fluorescence

scattering

label
antibody
antigens

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Problem: Fluor Emissions Overlap

- spectral compensation
- labor burden
- operating costs
- assay sensitivity
- limited # of channels

FITC

spectral spillover / crosstalk

PE
What If You Could Distinguish *Overlapping* Emissions?

**Intensity [%]**

- **FITC**
- **PE**

**Wavelength [nm]**

- Focus on one channel

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What If You Could Distinguish *Overlapping* Emissions?

**Fluorescence**

- **Lifetime:** SHORT → **PE**
- **Lifetime:** LONG → **FITC**

**Time** vs. **Fluorescence**

**Time** vs. **Wavelength**

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What If You Could Distinguish *Overlapping* Emissions?
**Time** Enables Discrimination of Overlapping Emissions

- **FITC** (long lifetime)
- **PE** (short lifetime)

Overlapping fluorescence (indistinguishable) vs. different lifetimes (distinguishable)
Traditional Flow Cytometry

- continuous light source
- one peak per event
- signal rises and falls as a cell passes through the laser beam
Kinetic River’s Secret Sauce: Time-Resolved Flow Cytometry

- modulated laser source
- hundreds of peaks per cell
- nanosecond decays captured
Patented Advantage: Time-Resolved Fluorescence Measurements

simultaneous measurement of multiple fluors with the same detector

excitation pulse

lifetime $\tau_1$

lifetime $\tau_2$
Features Unlocked by Kinetic River’s Time-Resolved Approach

- **Compensation-Free**
  - simplified protocols, lower costs of controls

- **Lifetime Tiers**
  - 2x – 3x parameters with same # of lasers, detectors

- **40+ Fluors**
  - like mass cytometry but cells can be sorted

- **Autofluorescence Removal**
  - lower background, higher sensitivity

- **Label-Free**
  - toward rapid cancer cell discrimination
Time-Resolved FC ➔ Compensation-Free FC

- no compensation

12-color platform using only
  - 2 lasers (405, 488 nm)
  - 6 fluorescence detectors
Time-Resolved FC $\Rightarrow$ 2x–3x # of Parameters

- each laser can support 3 lifetime tiers of fluors
  - 8–12 fluors *per laser*
  - 40+ fluors with 5 lasers
Time-Resolved FC \(\rightarrow\) Autofluorescence Removal

Fluorescence plus Autofluorescence

- Fluorescence signal with AF removed
- Stained Eos
- Unstained Eos

Fluorescence signal with AF removed

- Unstained Eos
- Stained Eos

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Kinetic River Key Facts

**Intellectual Property**
- 15 patents issued/allowed
- 13 patents pending
- 5 patent families

**Revenue Growth**

7-yr CAGR = 31%

**Key Customers**

- National Cancer Institute
- UCDavis

**Key Clients**

- National Research Council of Italy
- Stanford School of Medicine
- Sony
- STgenetics
- EMD Millipore
Kinetic River’s On-Market Products

Potomac

Shasta

Cavour

Tuolumne

Danube
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Expanding the Power of Flow Cytometry

• Time-resolved measurements eliminate bottlenecks in FC
• Reduce or eliminate spectral spillover, compensation
• Double to triple the number of detectable parameters
• Automatically remove autofluorescence background
• Label-free cancer cell discrimination

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Photonic Technology Needs

• **Sources**: pulsed OR high modulation frequency
  - pulsed: ~ 10 ns, ~ 10 MHz rep rate, ≥ 1 W peak power
  - modulation: 250+ MHz
  - from visible down to deep UV (266 nm)

• **Detectors**: compact, sensitive, high dynamic range
  - PMTs → APDs

• **Costs**: down, down, down
  - WW competition pushing down costs of analyzers
What Kinetic River Can Do For You

• **Vehicle to Market**: design your products into state-of-the-art technology from the very beginning

• **Market Intelligence/Strategy**: we provide consulting and training courses in flow cytometry to photonics companies interested in entering the space