



Delaware Flow NanoCytometer™:
Ultrasesitive Nanoparticle Detection in Flow

G. Vacca, A. Chin, K. LaneLutter, R. Hanson

NIH Flow Cytometry Interest Group 2023 Meeting
September 12, 2023

Delaware Flow NanoCytometer™



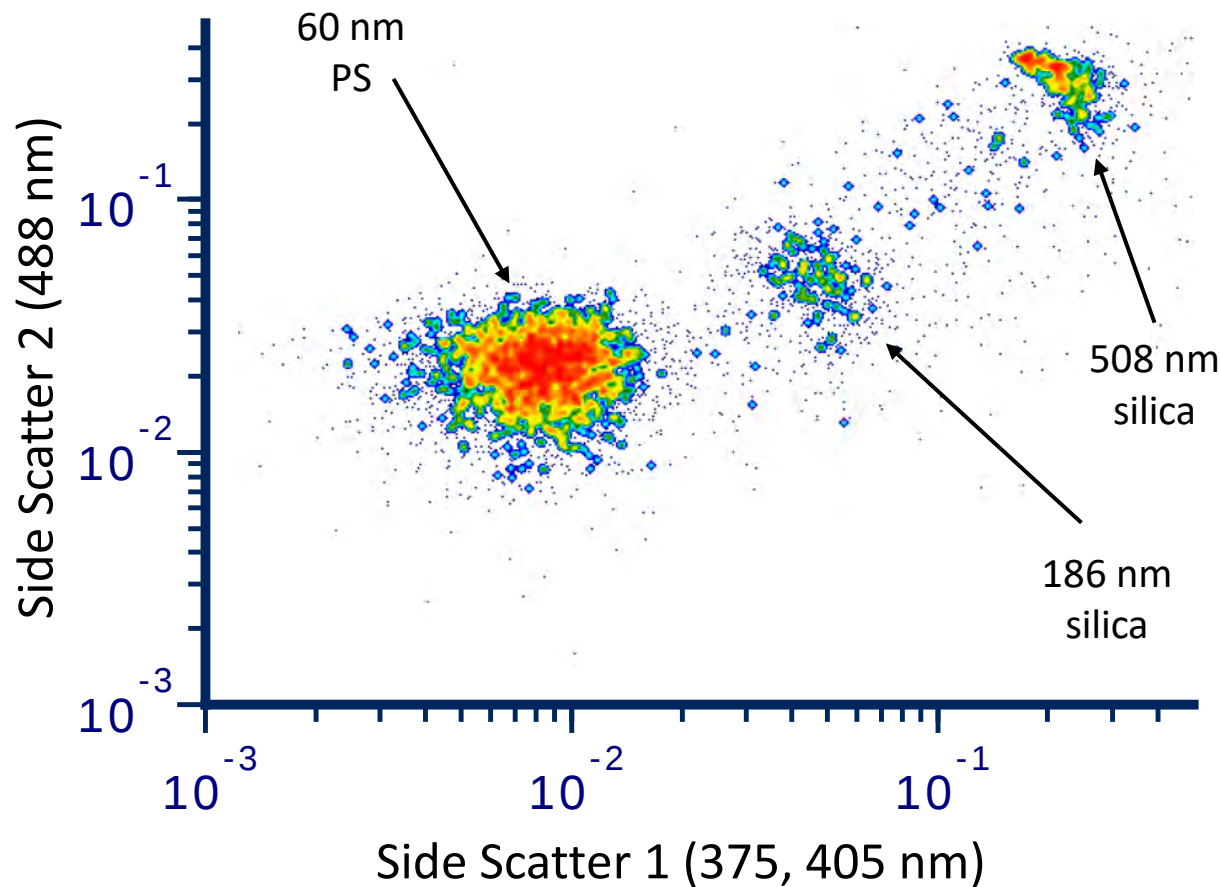
***Delaware* was built for purpose for EV researchers**

- high scattering and FL sensitivity
- two separate SSC channels (375 + 405, 488)
- did not compromise full functionality
- multiplexing of up to 6 FL channels
- high throughput (up to 2000 particles/sec)
- ease of use (including *Cavour*, *Shasta*, *Panama* acquisition and visualization software)
- not restricted to EVs, can do cell analysis also
- performance verified on a variety of materials

Delaware: 60-nm Scattering Detection Sensitivity (PS NPs)

Improve sensitivity

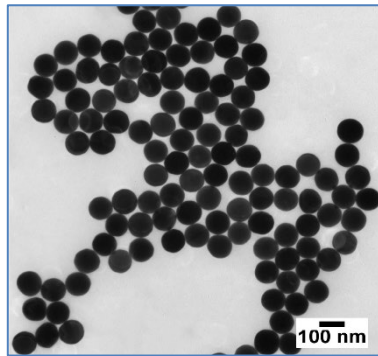
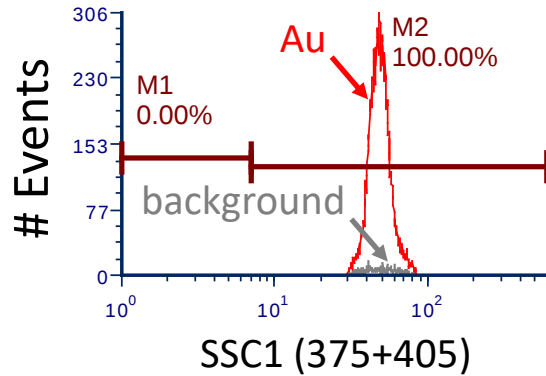
- low noise electronics
- proprietary collection optics
- high power lasers
- multiple wavelength scatter
- proprietary background reduction algorithms



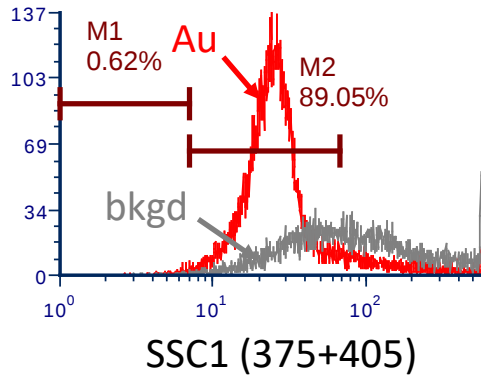
Silica nanospheres from Alpha Nanotech
Polystyrene nanospheres from Spherotech

Delaware: 28-nm Scattering Detection Sensitivity (Gold NPs)

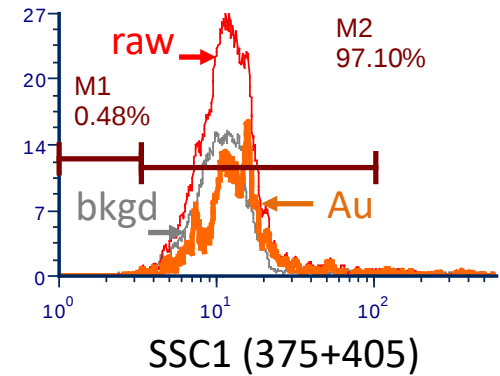
102 nm Gold



50 nm Gold

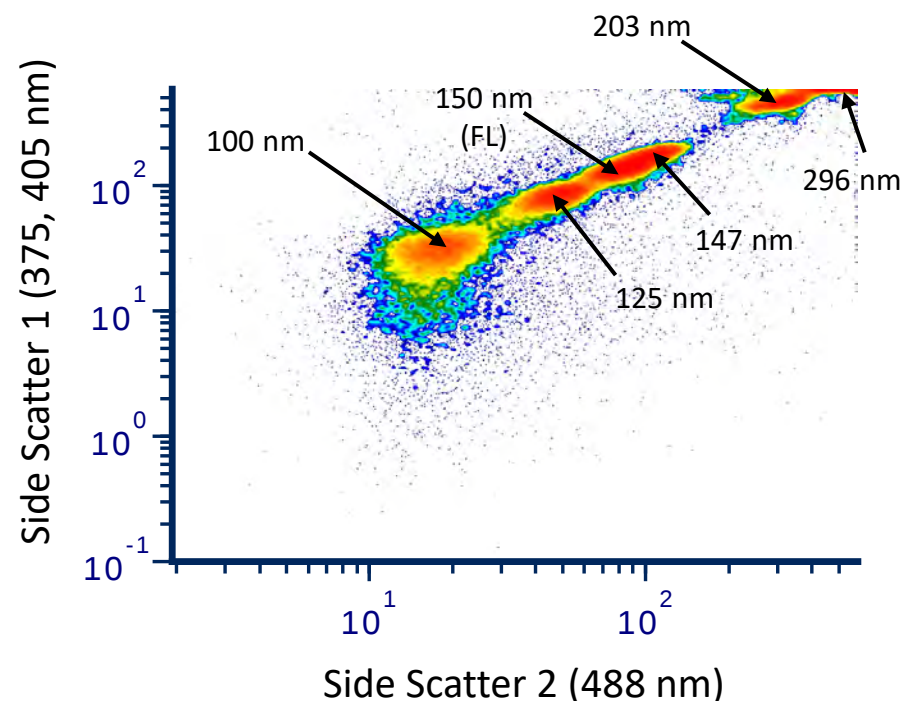
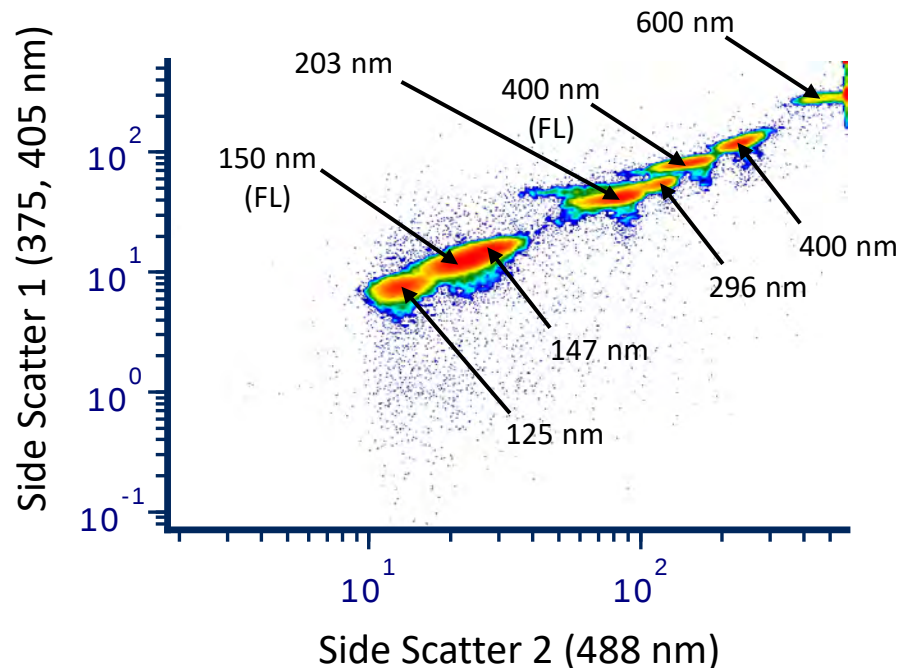


28 nm Gold

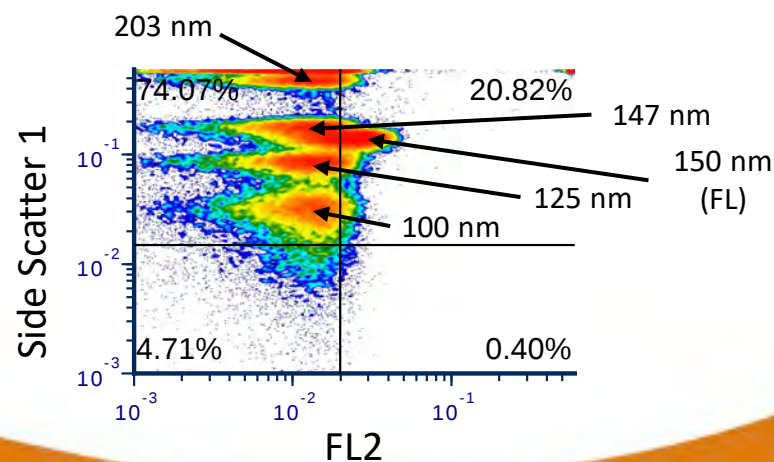


Gold NPs from nanoComposix
(sized by TEM)

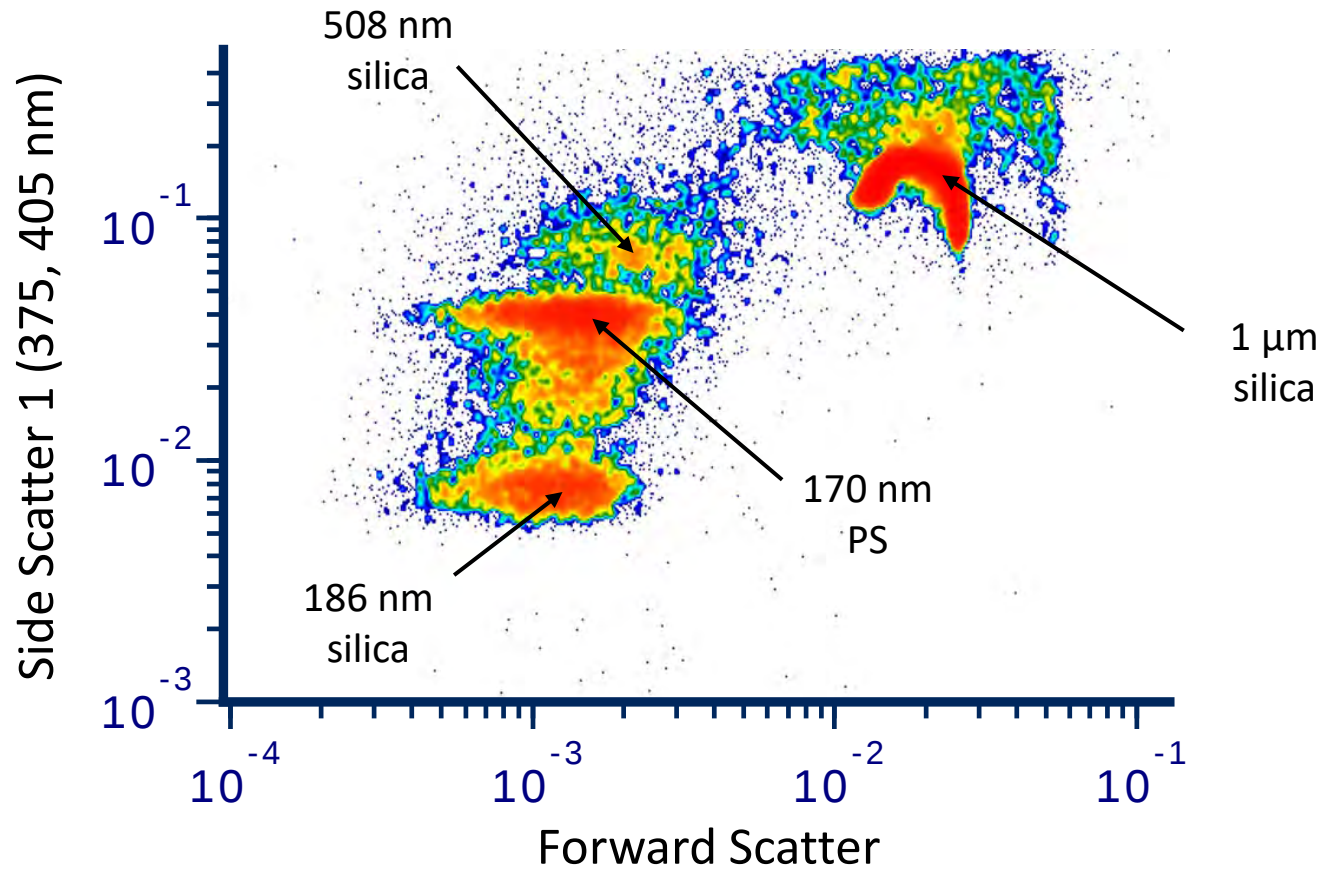
Delaware: 22-nm Resolution



Rosetta PS calibration
nanoparticles from Exometry

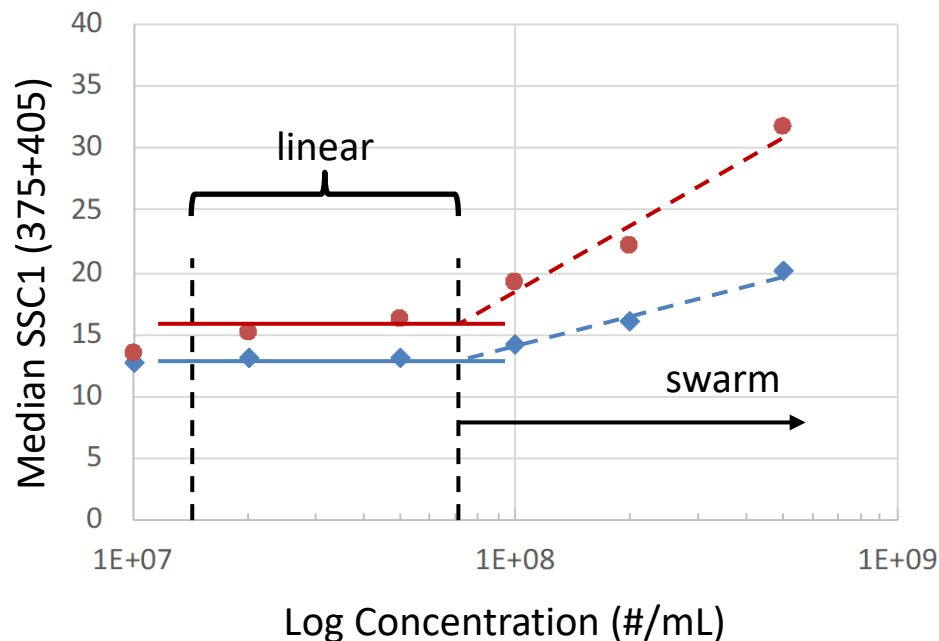
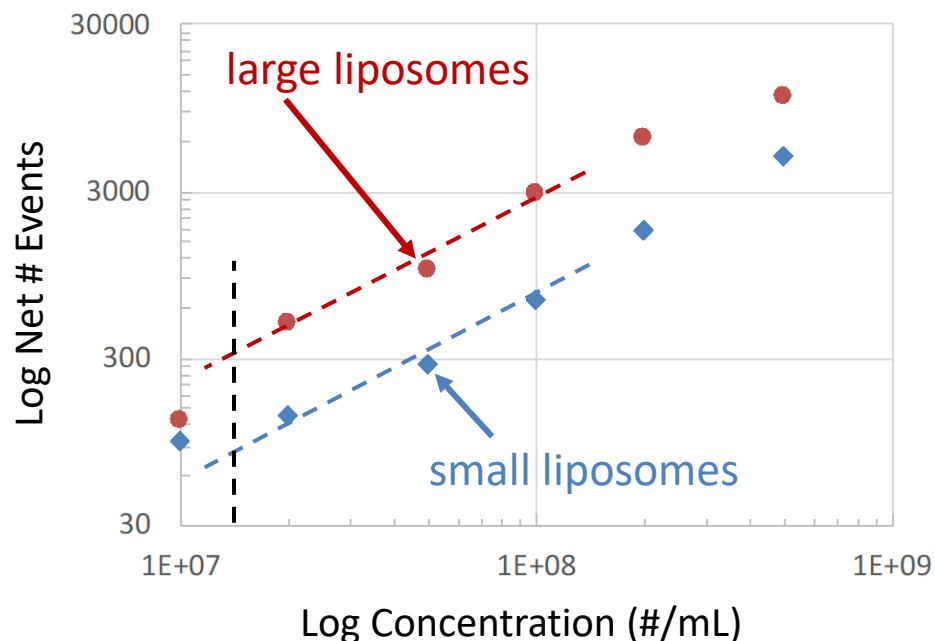


Delaware: Large Dynamic Range



Silica nanospheres from Alpha Nanotech
Polystyrene nanospheres from Spherotech

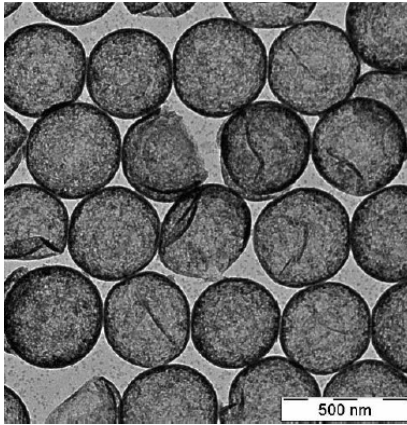
Delaware: Detection of 58- and 177-nm Liposome Standards



ACOERELA 
Acoerela liposome standards
Sized by nanoFCM at 177 nm and 58 nm

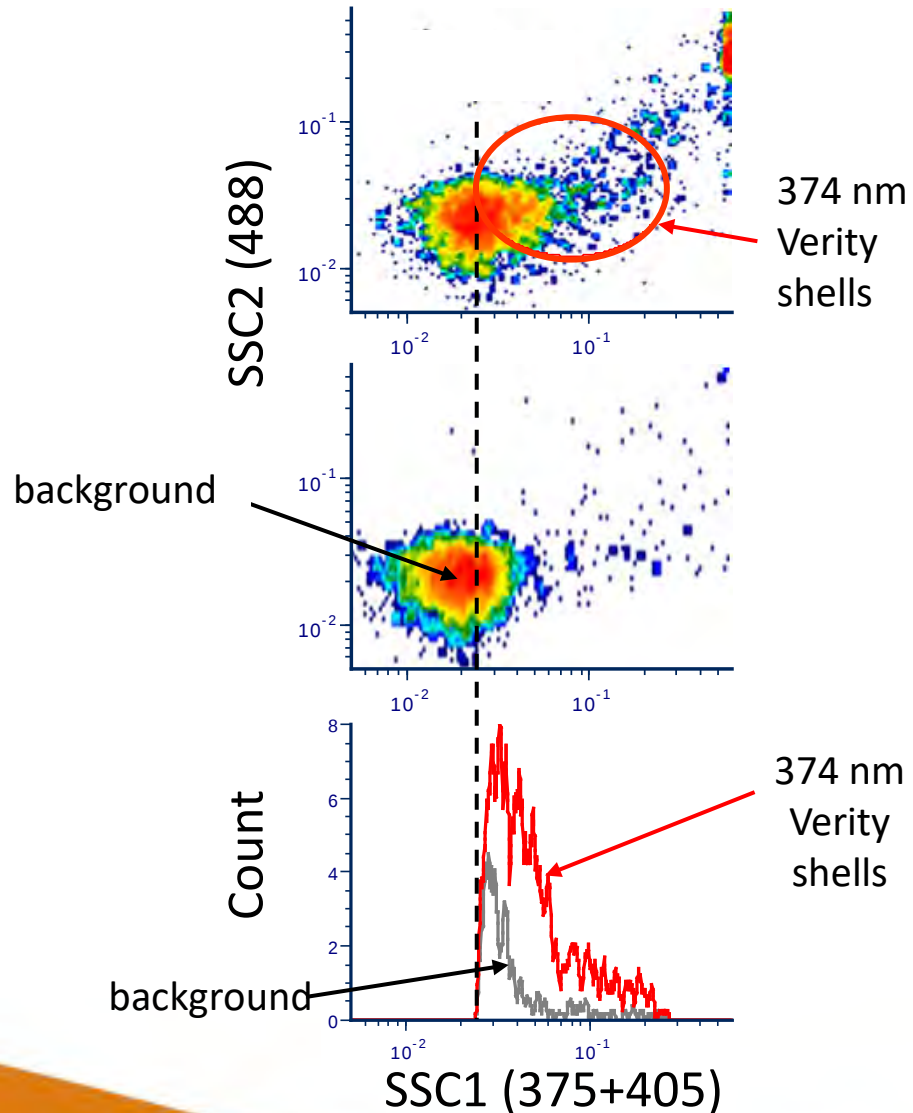


Delaware: Organosilica Shells

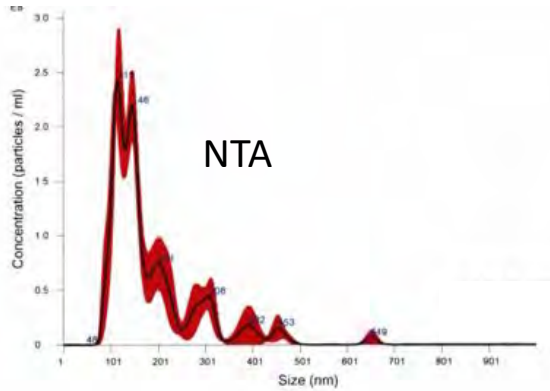


- very thin silica shell
- aqueous center
- very low scattering contrast

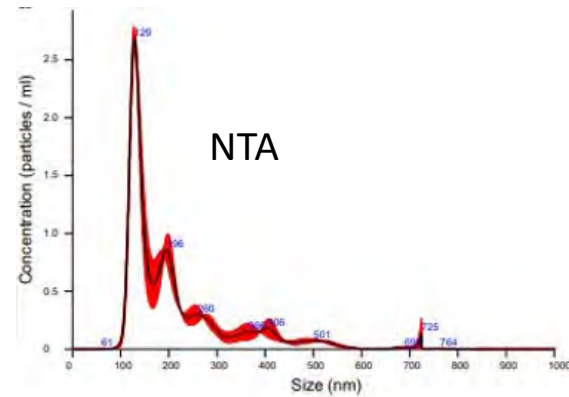
374-nm Verity Shells™ hollow organosilica nanosphere shells from Exometry



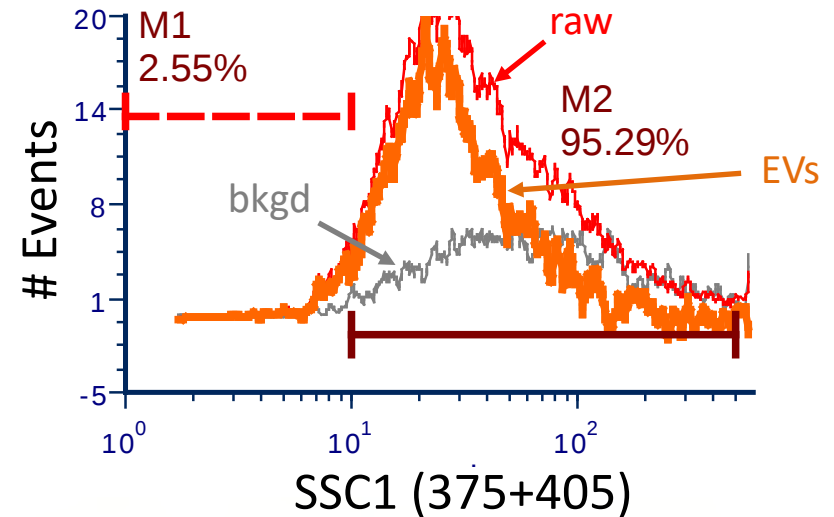
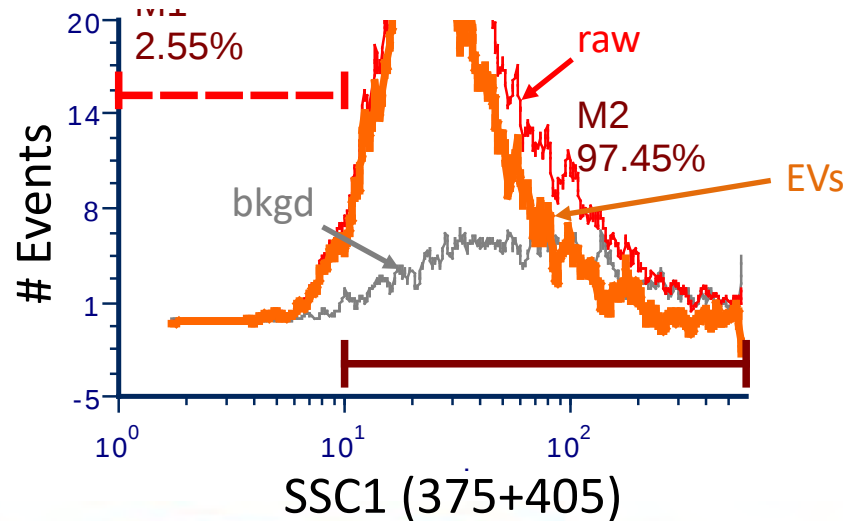
Delaware: HeLa- and PC3-Derived EVs



HeLa EVs



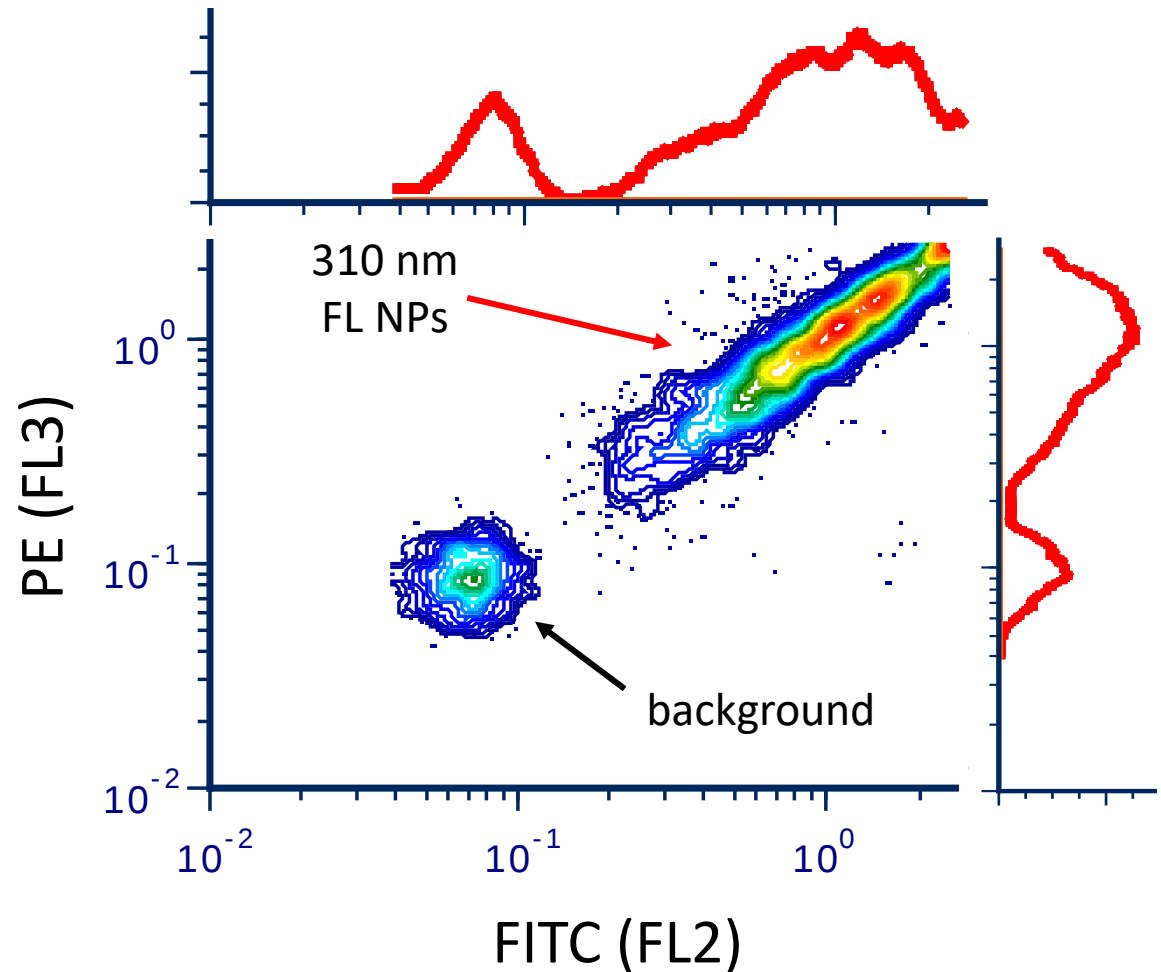
PC3 EVs



EVs purified and measured on NTA in the lab of Dr. Fatah Kashanchi at George Mason University

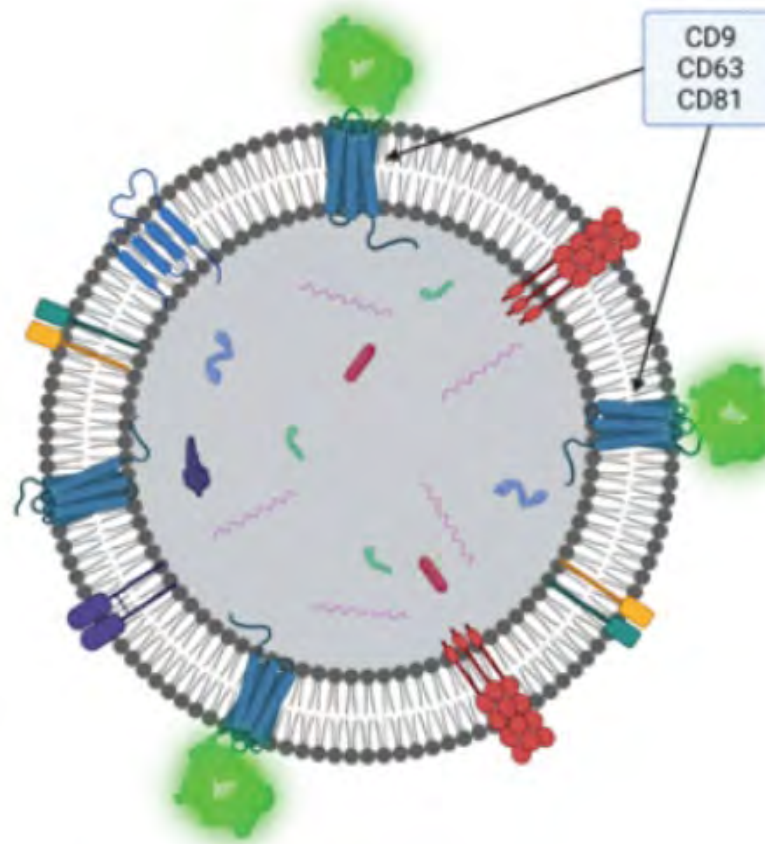
Delaware: Up to 6 Fluorescence Channels

- up to 6 FL channels
- analysis of cargo
- cross-correlate marker expression in individual EVs
- critical for correlating with EV function

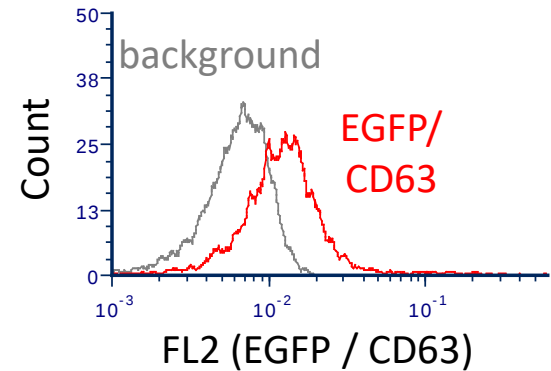
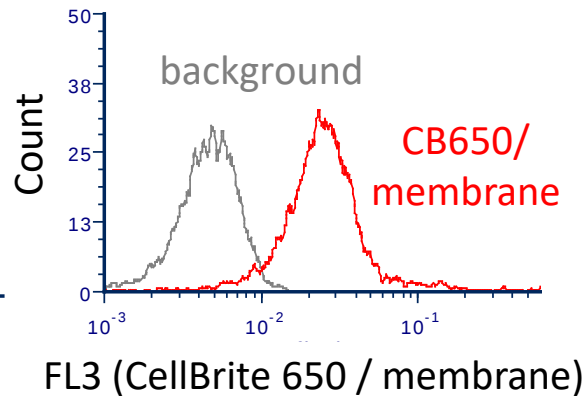
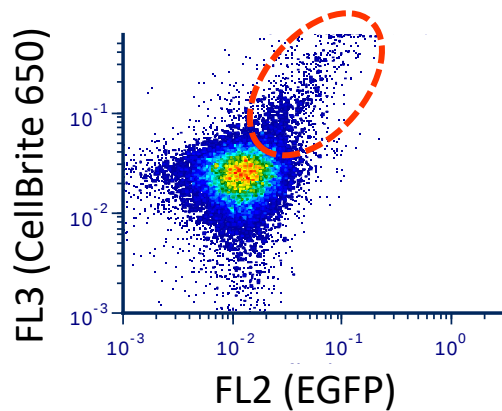
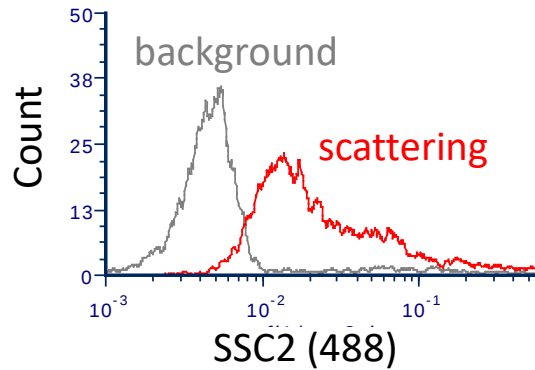
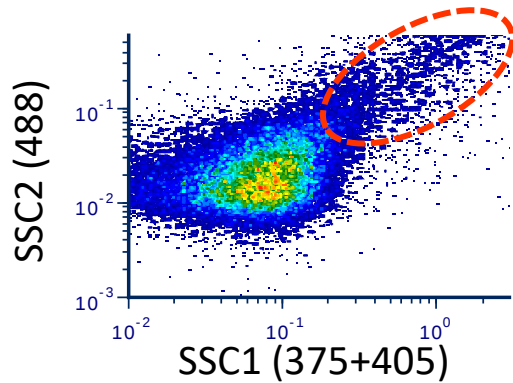


Spherotech UltraRainbow 310-nm fluorescent nanoparticles

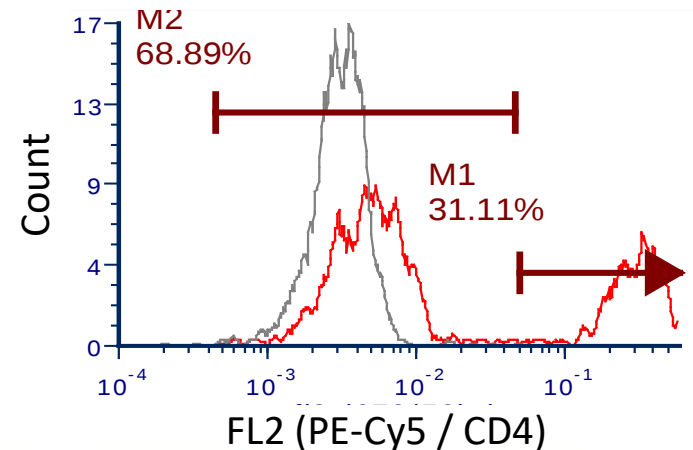
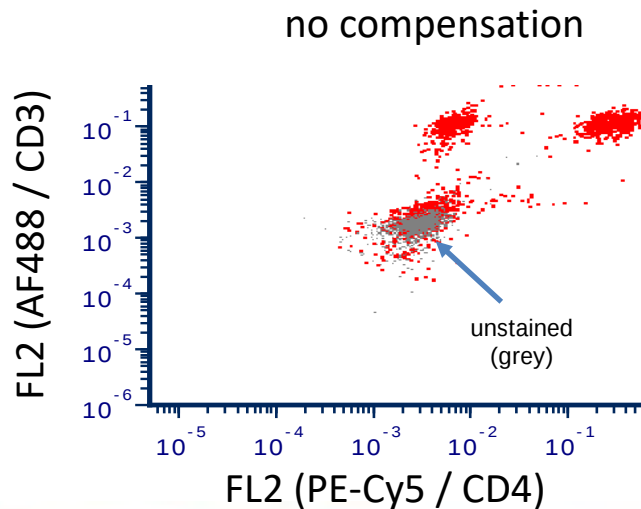
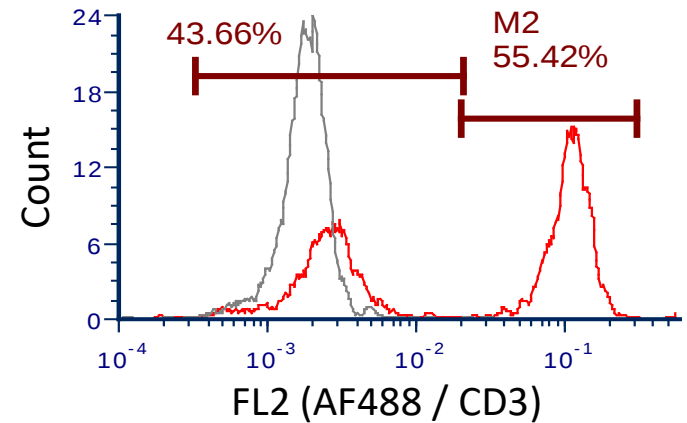
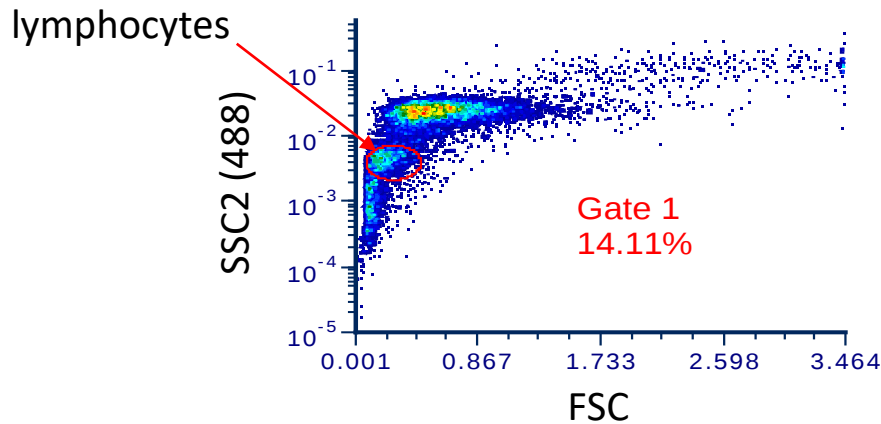
Delaware: Tetraspanin Detection in EVs



Delaware: Tetraspanin Detection in EVs



Delaware: Stained VeriCell™ Leukocytes



Veri-Cell PBMCs are from BioLegend and were stained for CD3 and CD4 in-house

Delaware Configurations

Basic Configuration	High Sensitivity Configuration	Five-Laser Configuration
2 Lasers	3 Lasers	5 Lasers
405 nm, 250 mW 488 nm, 200 mW	375 nm, 70 mW 405 nm, 250 mW 488 nm, 200 mW	375 nm, 70 mW 405 nm, 250 mW 488 nm, 200 mW 561 nm, 100 mW 640 nm, 150 mW
Standard Scattering	Ultrasensitive Scattering	Ultrasensitive Scattering
FSC, SSC (405 and 488 nm)	FSC, SSC (375, 405, 488 nm)	FSC, SSC (375, 405, 488 nm)
2 Fluorescence Channels	4 Fluorescence Channels	6 Fluorescence Channels
525/50 580/23	525/50 580/23 615/24 697/58	440/40 (optional) 525/50 580/23 615/24 697/58 755/35

Delaware Flow NanoCytometer™ for EV Analysis

- 3 configurations for broad range of needs
 - up to 5 lasers
 - fluorescence detection in up to 6 channels
- 60-nm single-NP detection (PS)
- 28-nm single-NP detection (Au)
- 58-nm single-liposome detection
- 22-nm resolution
- cancer cell EVs
- tetraspanin detection in HEK293 EVs
- low background
- wide dynamic range
- intuitive user interface
- *also* measures cells



KineticRiver.com/Delaware
info@KineticRiver.com

The *Delaware* provides excellent size sensitivity and resolution **without compromising** multiplexing, throughput, or ease of use



QUESTIONS?

