



2° European Biosensor Symposium 2019

EBS2019

Florence, Italy, February 18-21, 2019



Microarray based platform for extracellular vesicles imaging and phenotyping

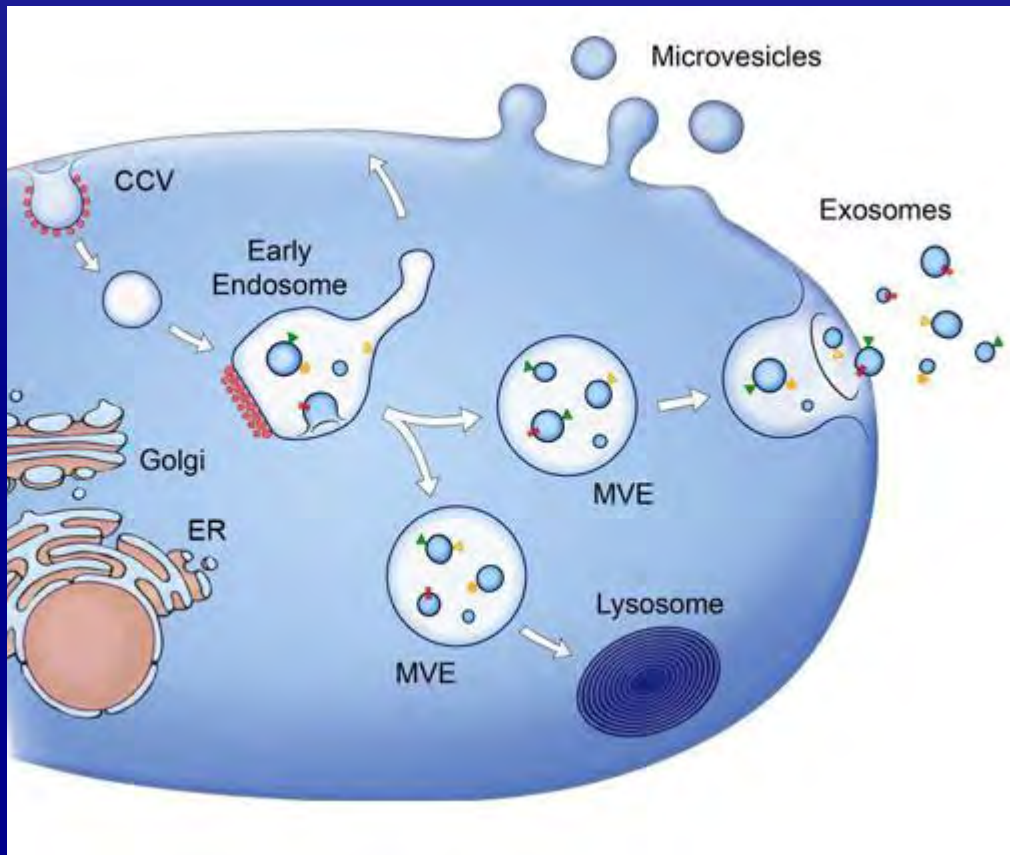
D. Brambilla, M. Cretich, A. Romanato, M. Odinolfi, E. Chiodi,
S. Descroix, A. Yalcin Ozkumur, M.S. Unlu, M. Chiari



- ✓ **Microarray platform for Exosome Immune-Capture**
- ✓ **Interferometric detection of Single NanoParticles**



Extracellular Vesicles

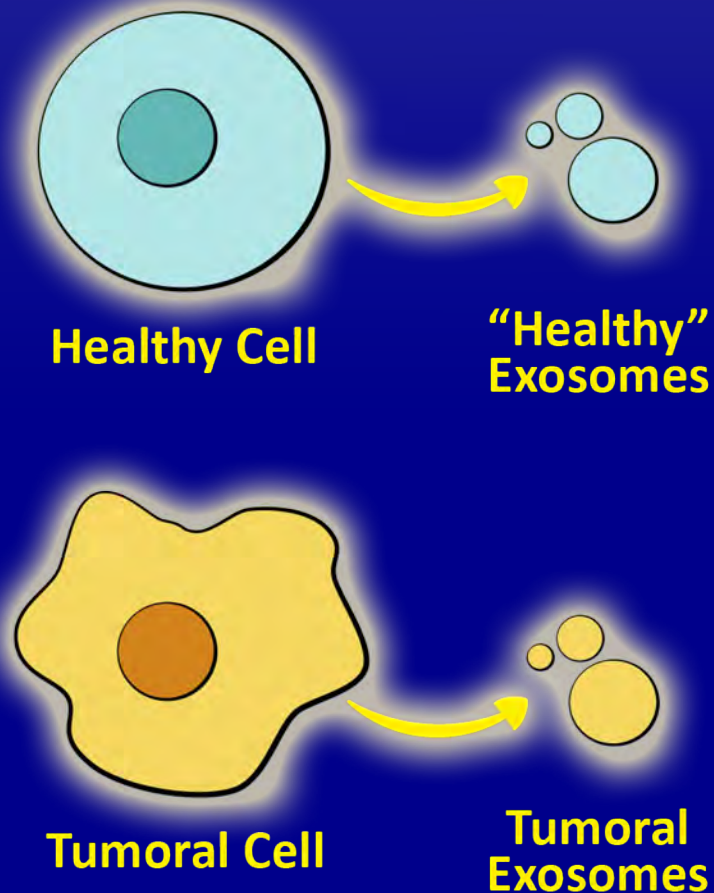


Exosomes

- ☐ Early Endosome
- ☐ Multi Vesicular Bodies
- ☐ Lysosomal degradation
- ☐ **Exosome Release**



Exosome: Diagnostic potential



☐ Mirrors the cell of origin

☐ **Liquid Biopsy**

✓ Cancer

✓ Neurodegenerative diseases

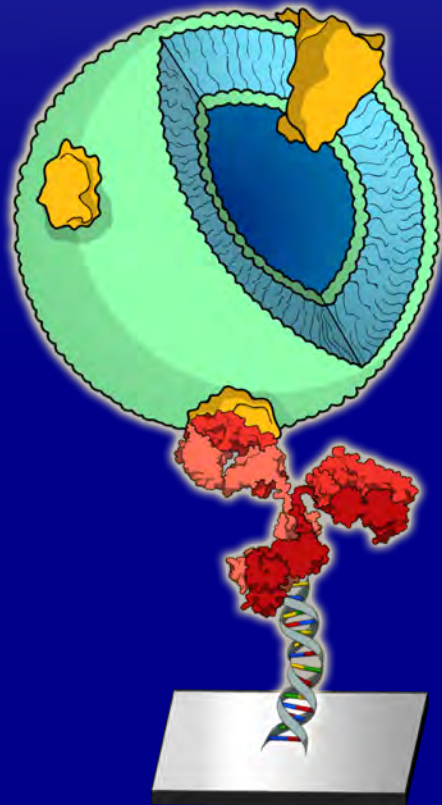


Exosome: Characteristics

- ✓ Number
- ✓ Size distribution
- ✓ Membrane composition
- ✓ Cargo



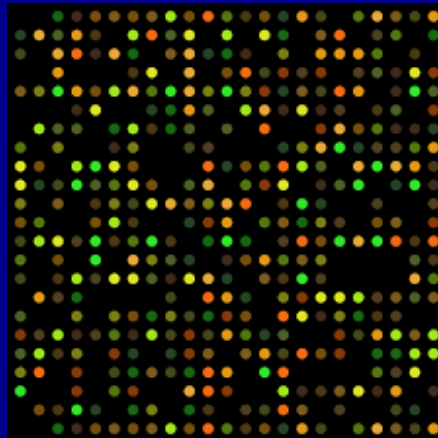
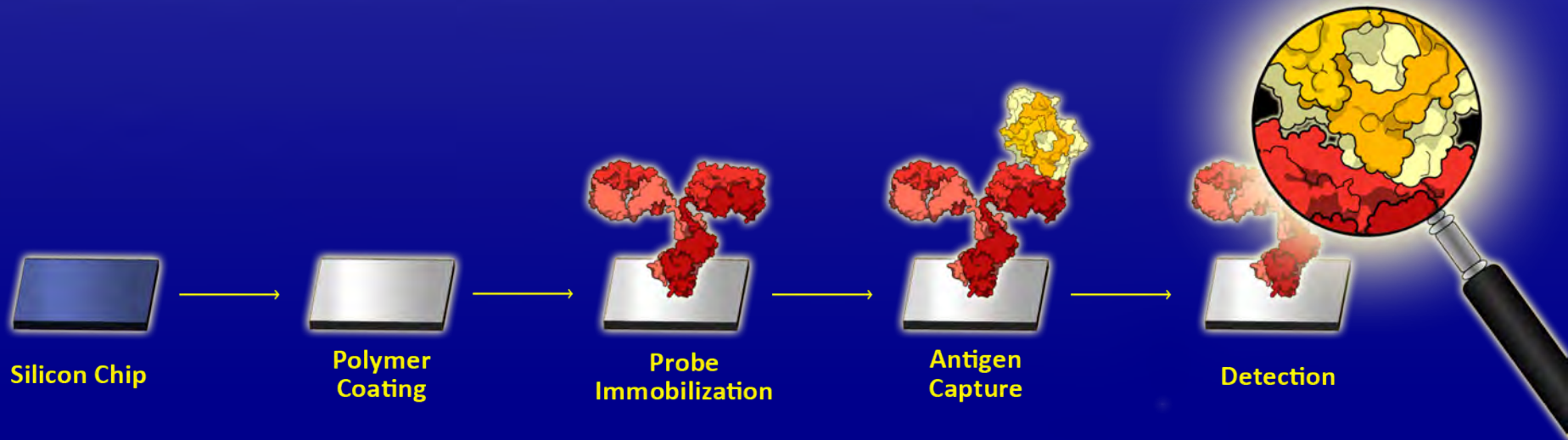
Exosome: Capture



✓ Microarray platform for
Exosome Immune-Capture



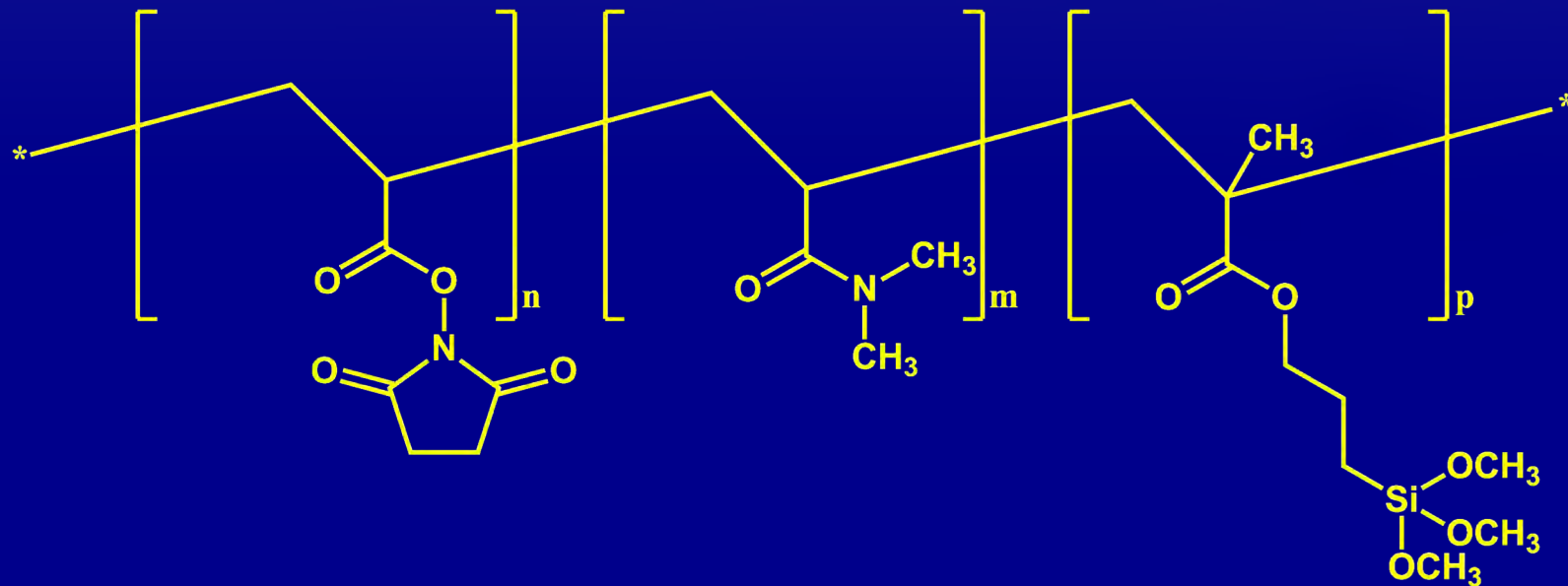
Microarray technique





Polymeric Coating

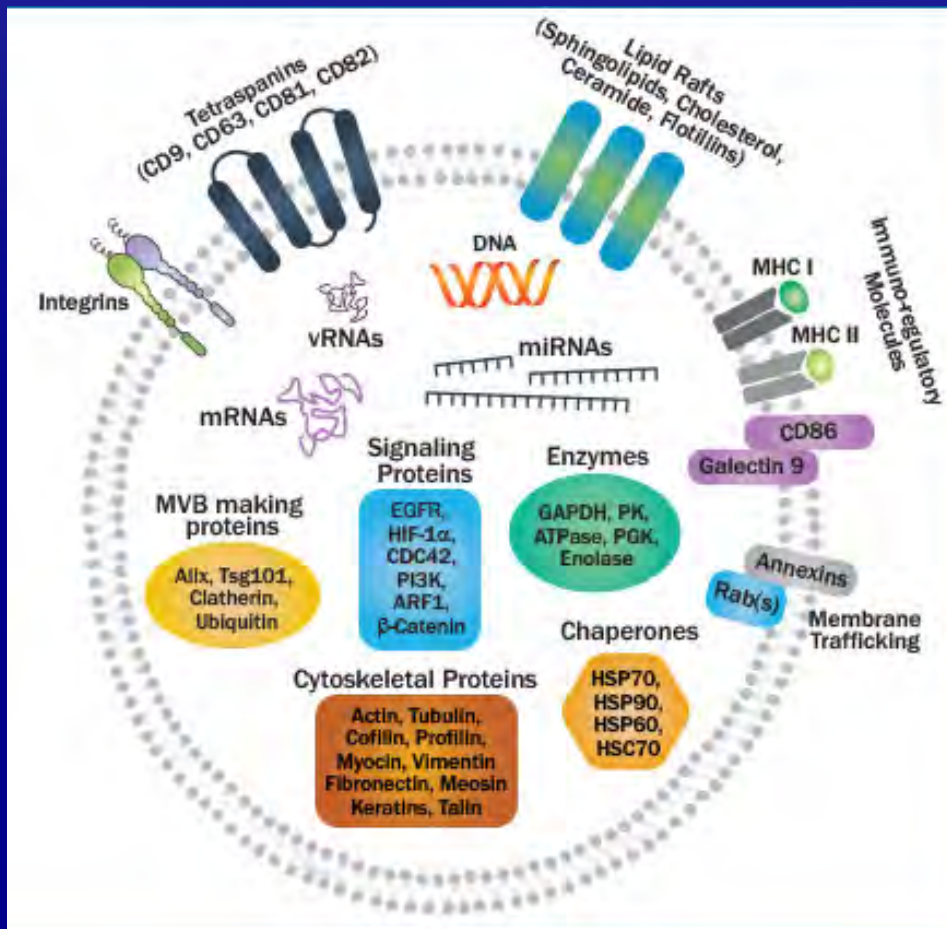
Copoly(DMA-NAS-MAPS)



m=97
n=2
p=1



Probe Immobilization

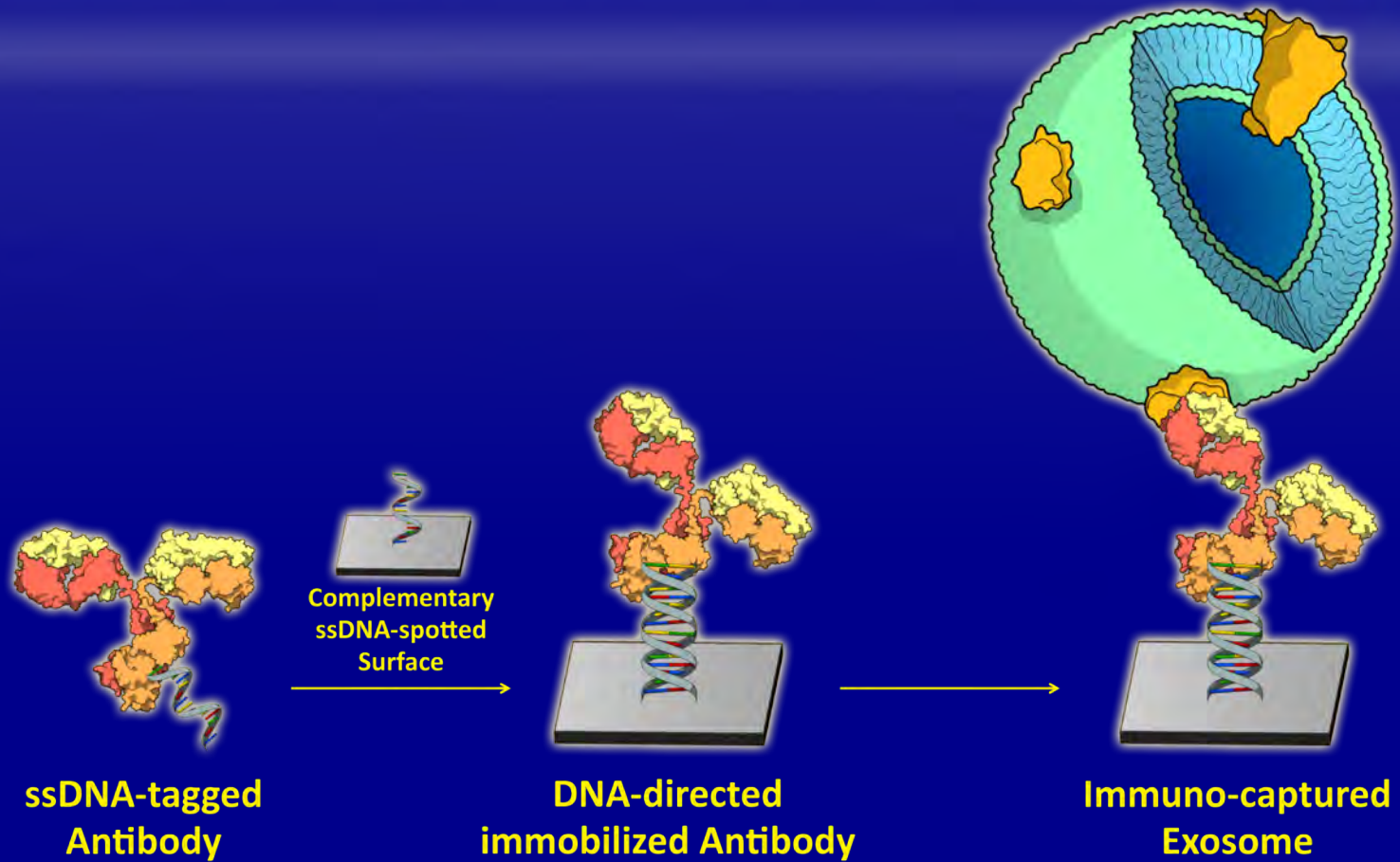


Antibodies (proteic probes)

✓ **Tetraspanins (CD9, CD63)**

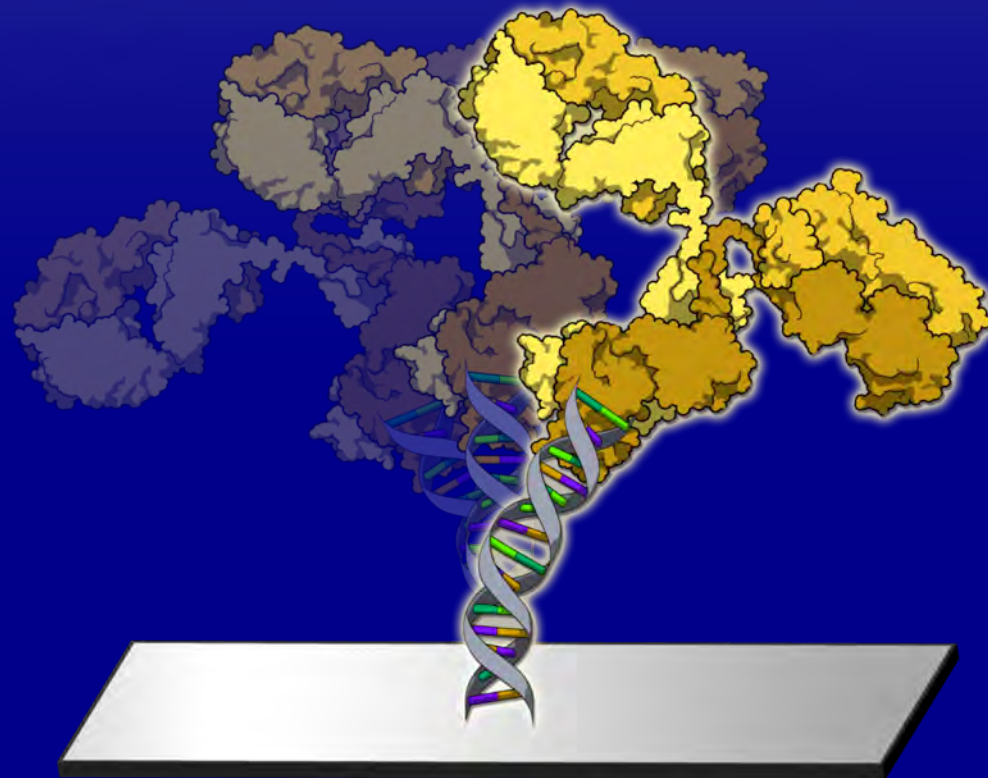


DNA-directed Immobilization #1



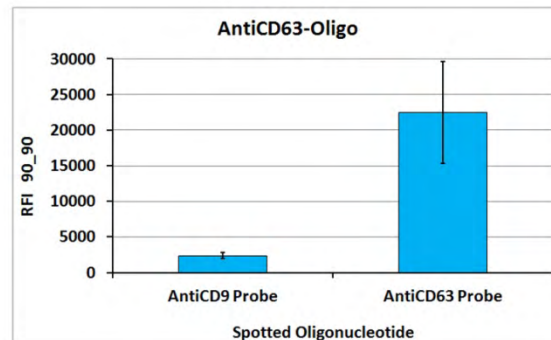
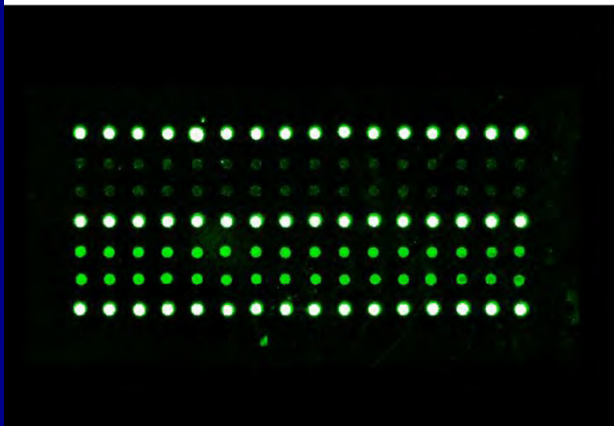
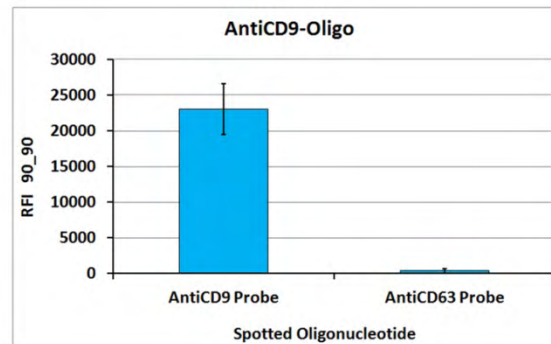
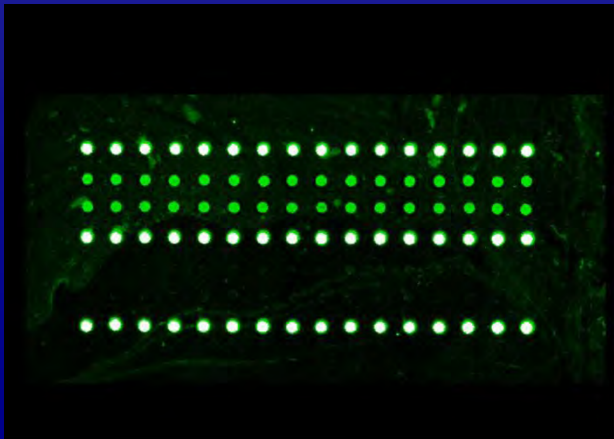


DNA-directed Immobilization #2



- ✓ DNA Spot morphology
- ✓ Antibody specificity
- ✓ **Improved flexibility**

Selectivity



✓ Multiplexing

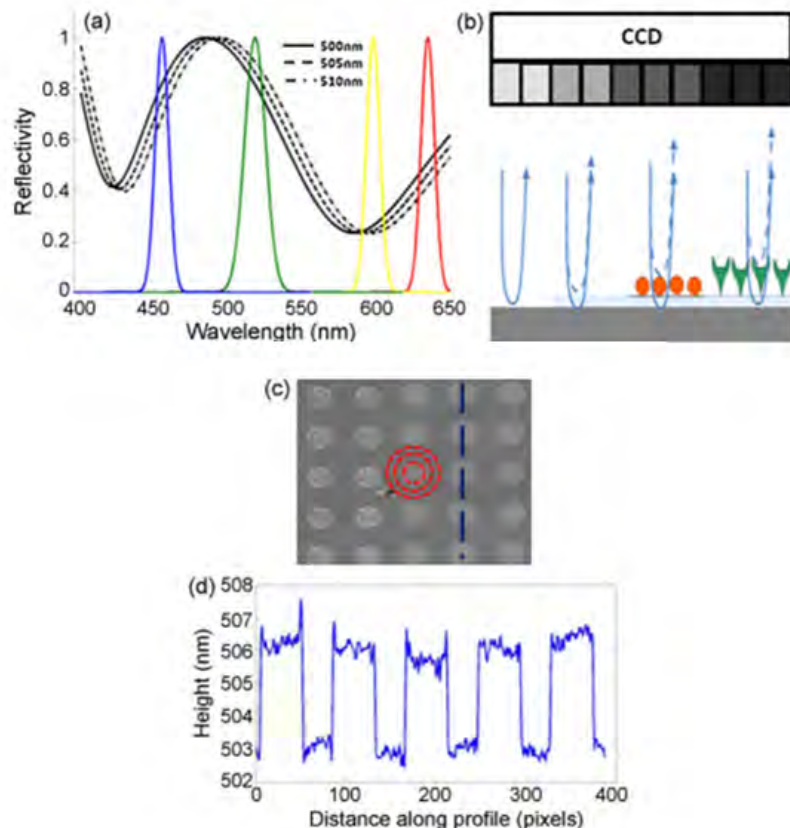


Detection

- ✓ Interferometric detection of Single NanoParticles



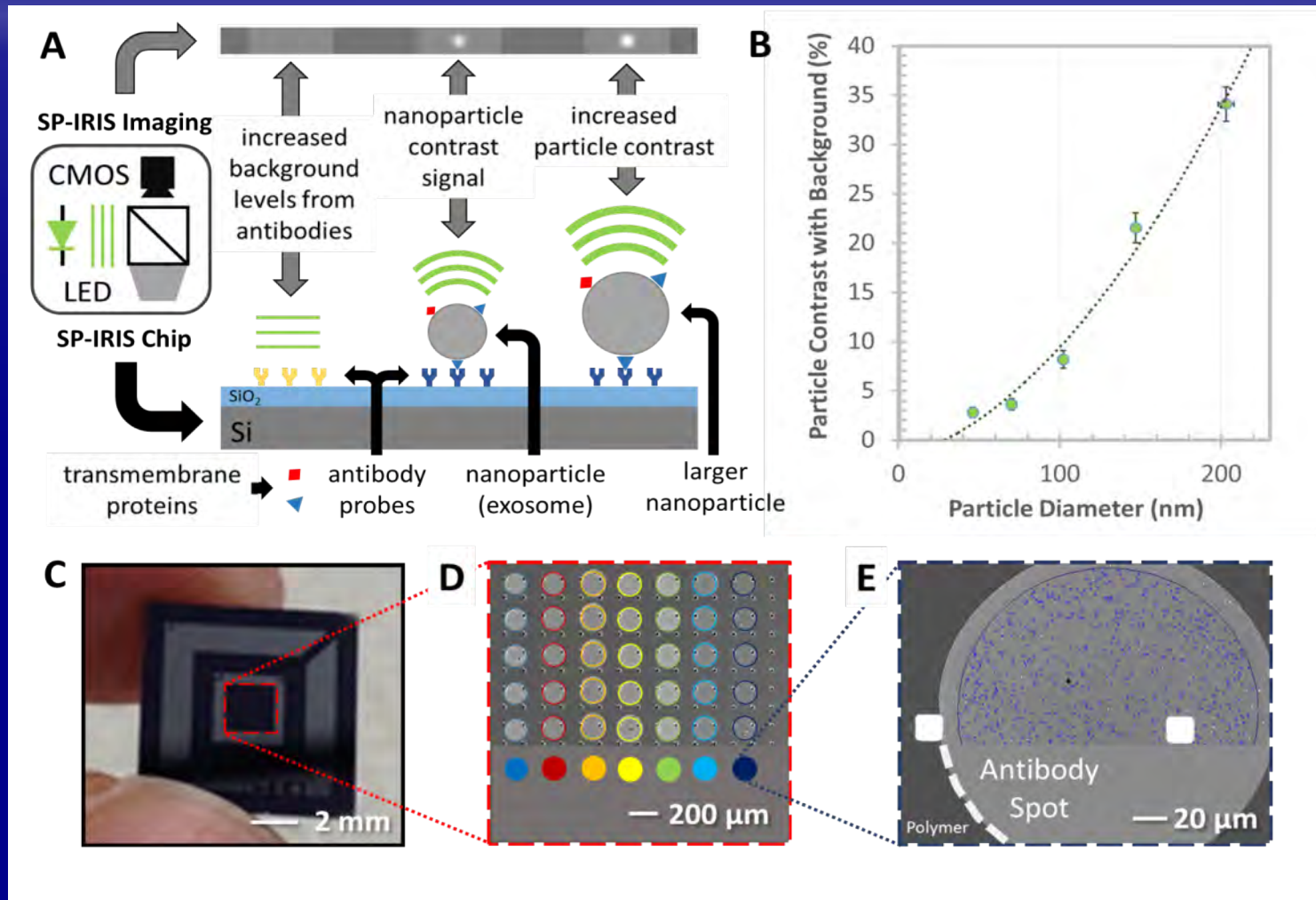
SP-IRIS: Sizing and Phenotyping #1



✓ IRIS: Interferometric Reflectance Imaging System

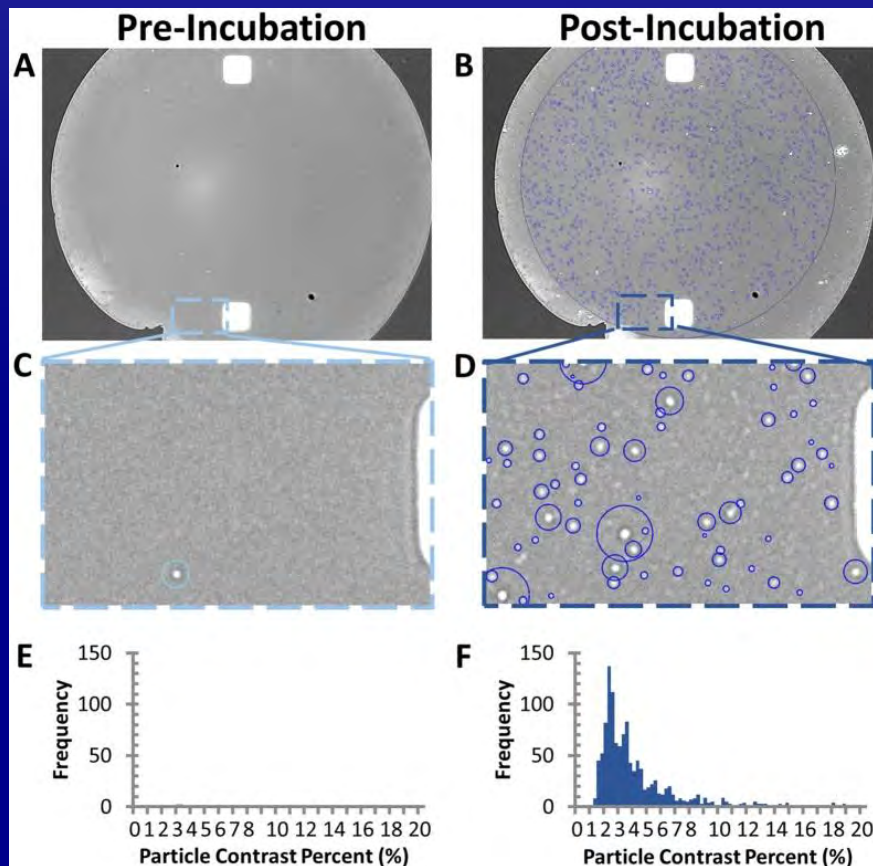
- Led-based technique
- Matter immobilized on surface
- Shifts of reflectivity
→ Image contrast

SP-IRIS: Sizing and Phenotyping #2





SP-IRIS: Sizing and Phenotyping #3



- ✓ **Digital count** of nanoparticles (50-200 nm)
- ✓ **Size distribution**
- ✓ **Phenotyping** (microarray platform)



Exosome Immune-capture: Results #1

pre incubation

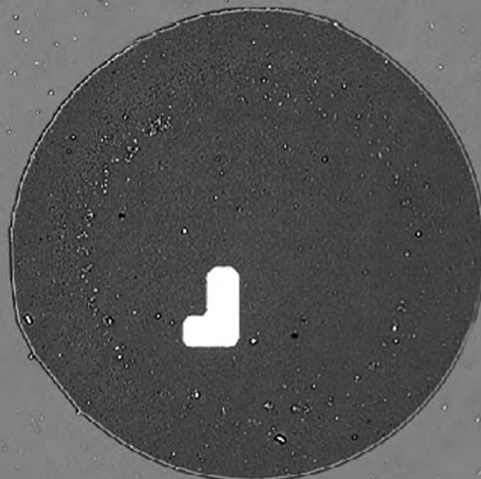
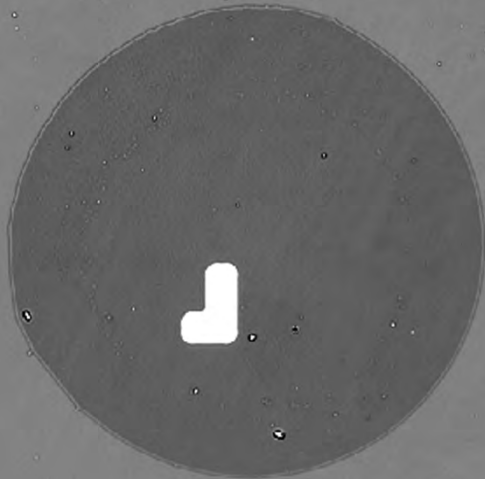
post incubation

Spot:

DNA-directed
antiCD9

Incubation:

Human Blood
Serum,
2.5h, RT





Exosome Immune-capture: Results #2

pre incubation

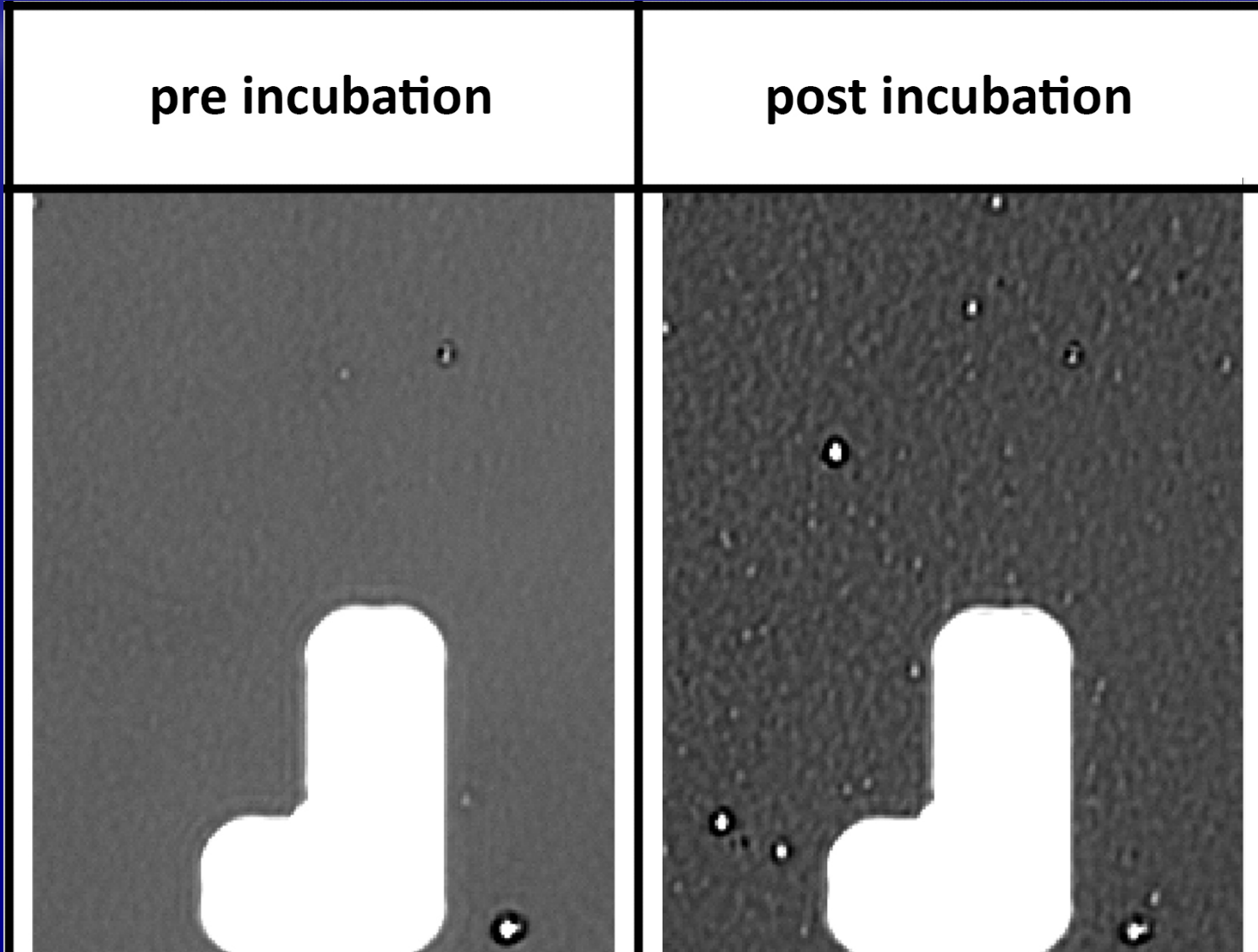
post incubation

Spot:

DNA-directed
antiCD9

Incubation:

Human Blood
Serum,
2.5h, RT





Exosome Immune-capture: Results #3

pre incubation

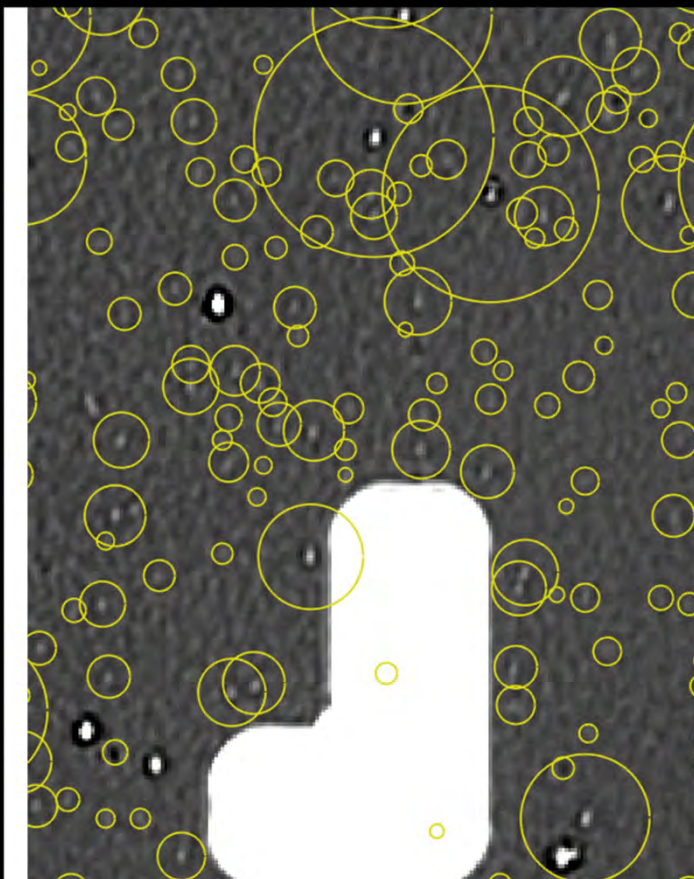
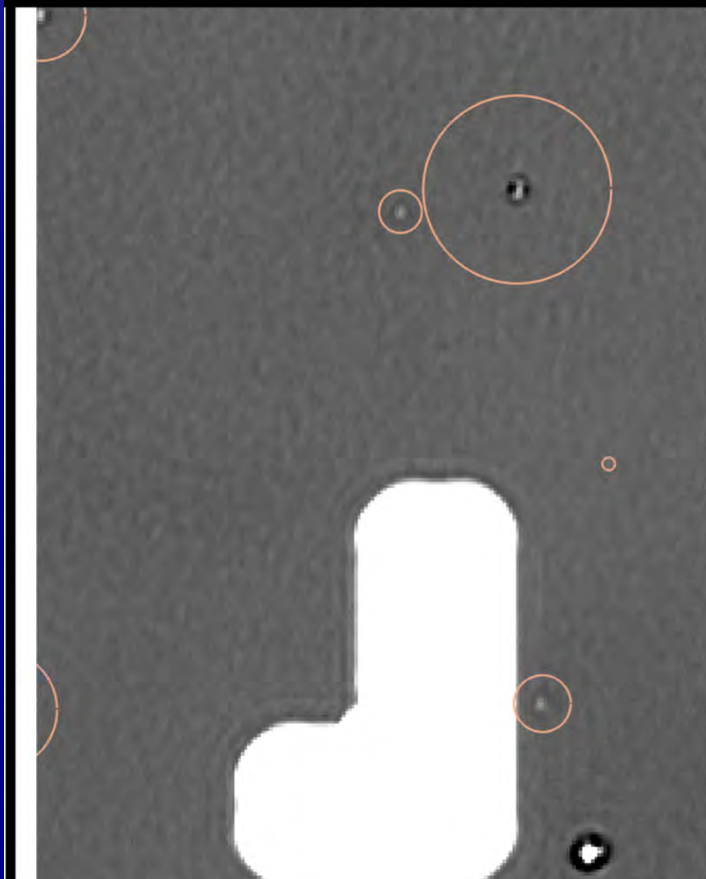
post incubation

Spot:

DNA-directed
antiCD9

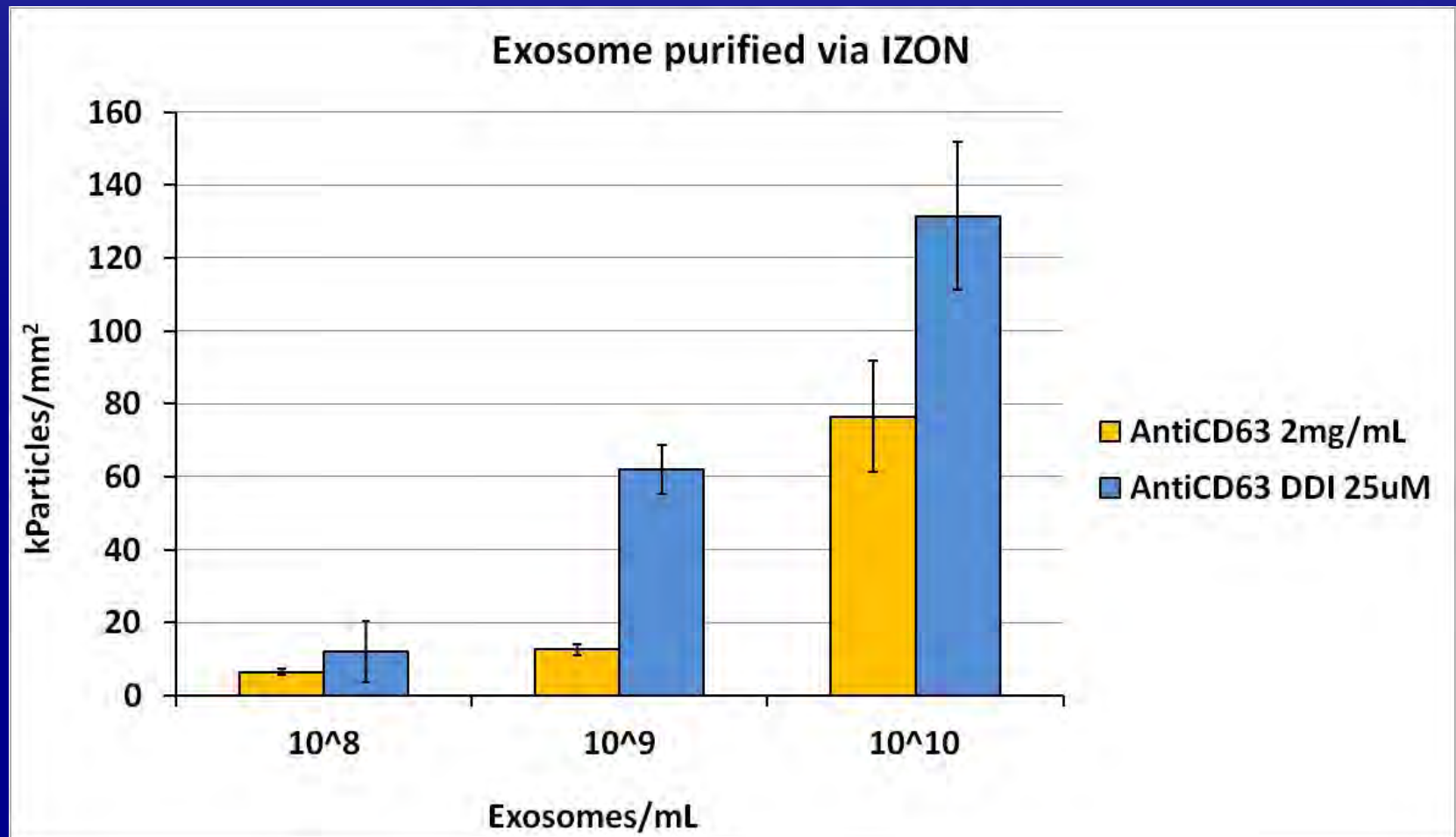
Incubation:

Human Blood
Serum,
2.5h, RT

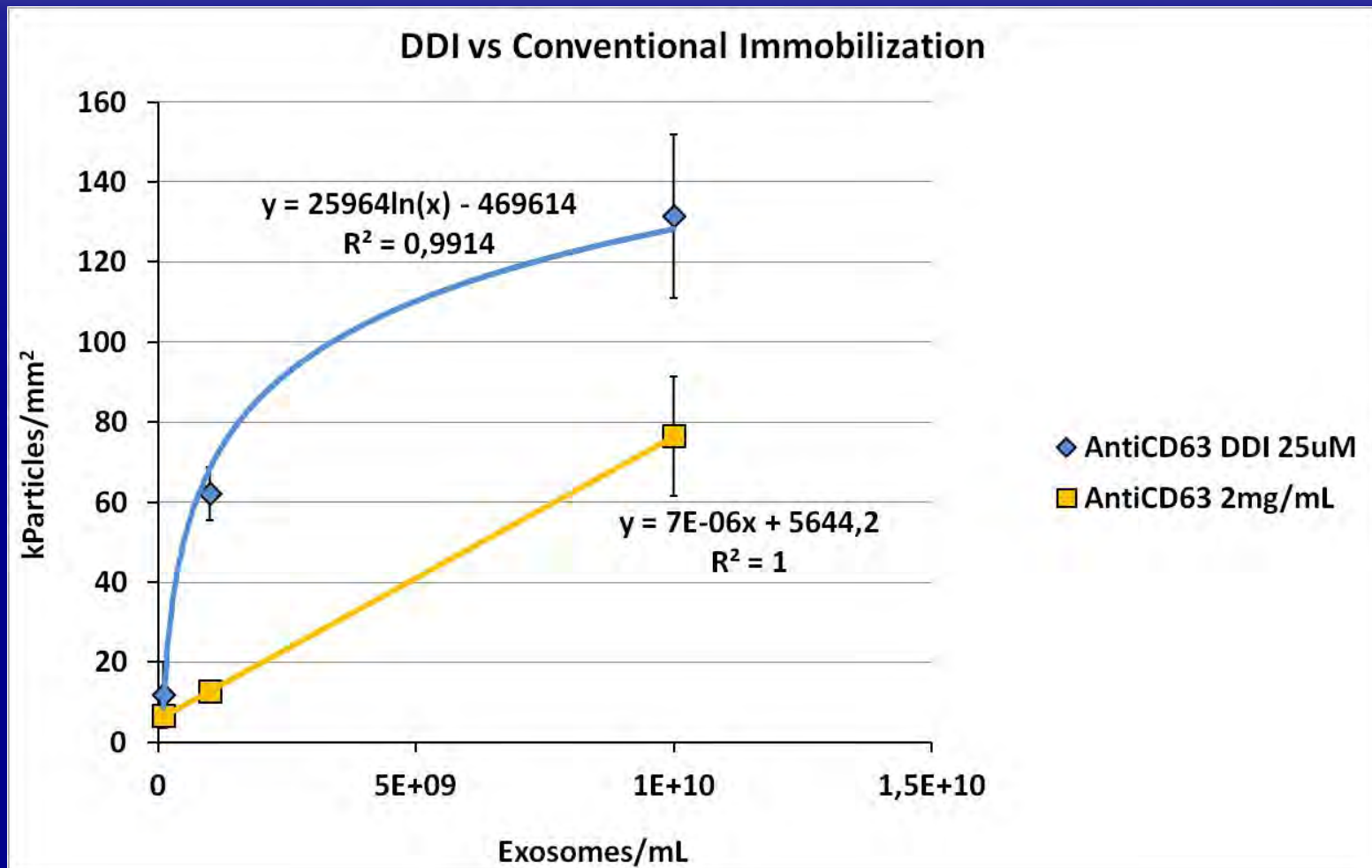




Exosome Immune-capture: Results #4



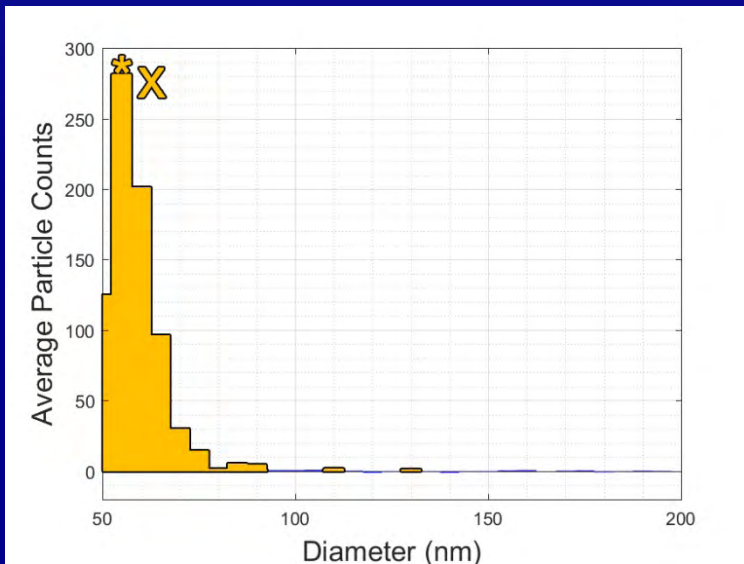
Exosome Immune-capture: Results #5



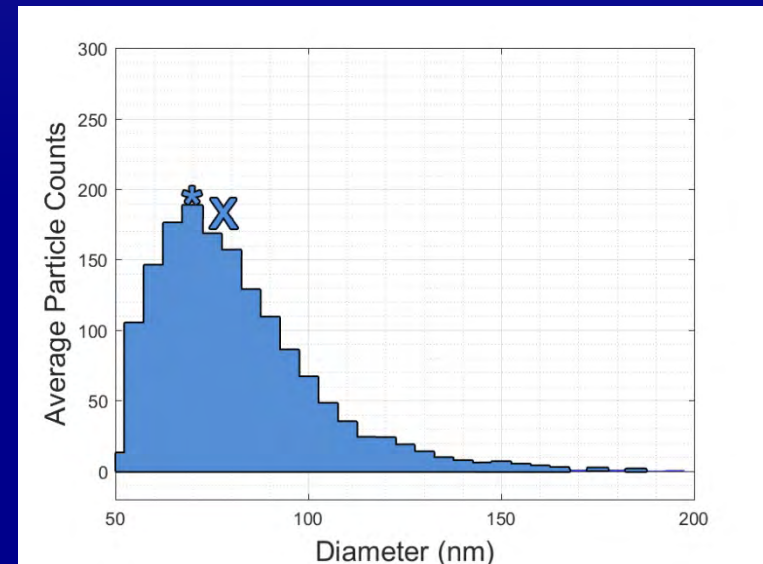


Exosome Immune-capture: Results #6

unmodified AntiCD63



DNA-Directed AntiCD63





Next Step: Real Time SP-IRIS

- ✓ K_d constant \longrightarrow **Affinity**
- ✓ Distinguish specific and non-specific events



Acknowledgments



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indEx

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**THANK YOU FOR
THE KIND
ATTENTION**