Interferometric Reflectance Imaging Sensor using Si-based Microfluidics

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Electrical Engineering,

Physics,

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Graduate Medical Sciences

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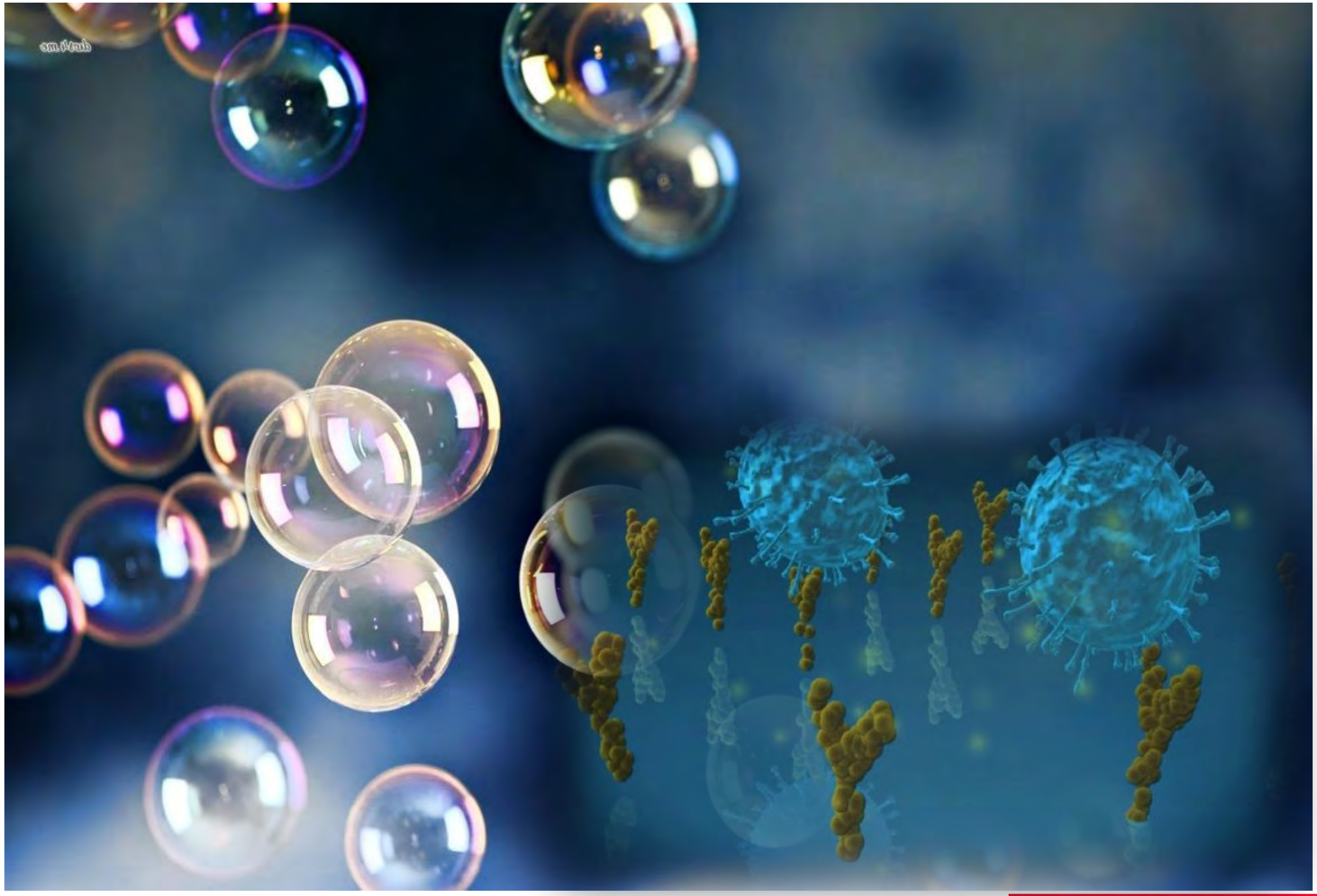

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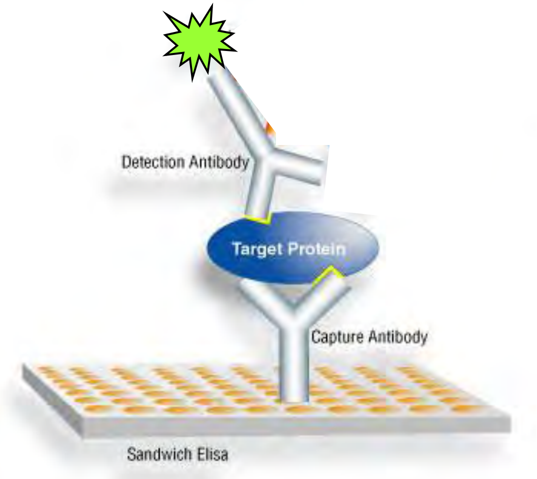
Outline

- Motivation
- Interferometric Reflectance **Imaging** Sensor
- **Sensor Chip and Cartridge Development**
- Versatile platform
  - Molecular kinetics
  - Biological Nanoparticle Detection and Sizing
  - Resolution improvement by oblique illumination and reconstruction
  - Digital microarrays
- Conclusions and Future

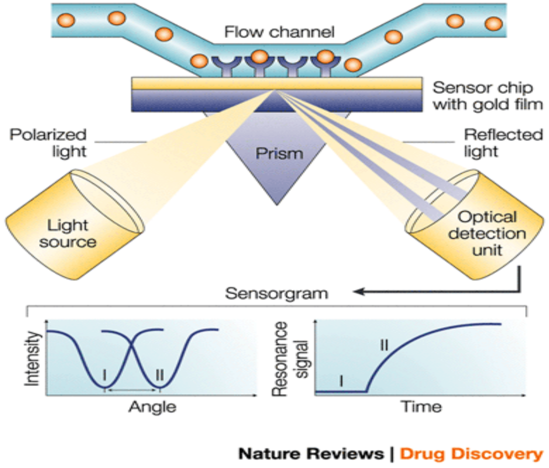
Motivation - Diagnostics/detection

ELISA
(Enzyme-linked immunosorbent assay)



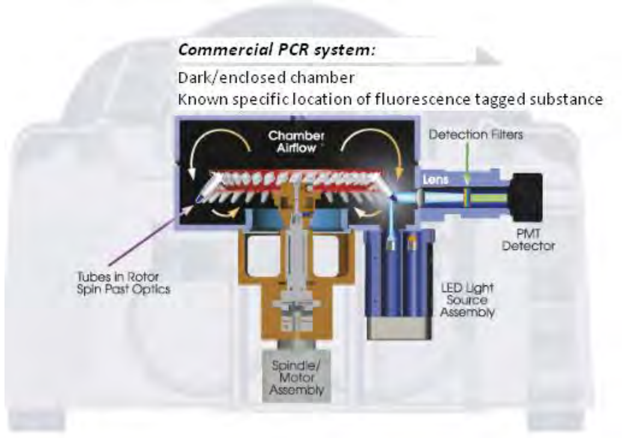
- Uses secondary labeling for detection
- Requires laboratory and skilled technicians
- Time-consuming process

SPR
(Surface Plasmon Resonance)

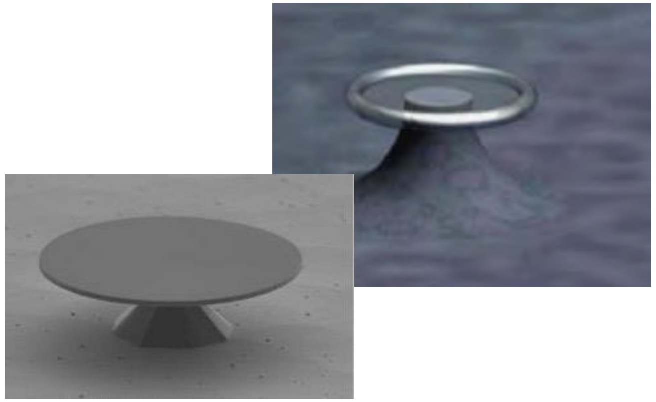


- Label-free optical detection
- expensive disposables
- Au surface

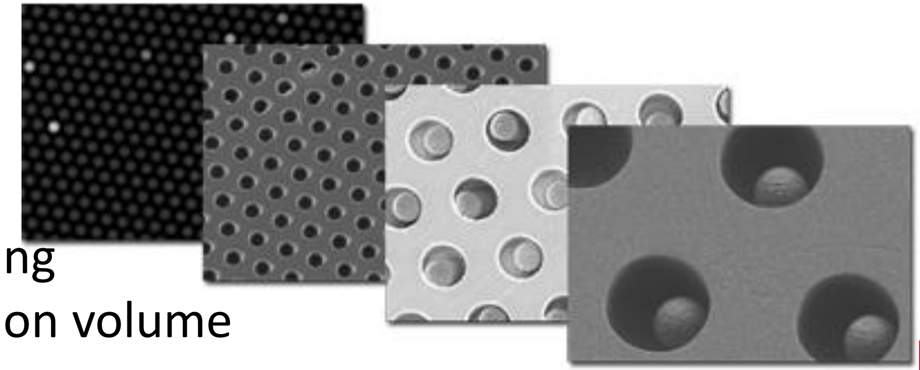
PCR
(polymerase chain reaction)



- Sample preparation,
- Dark/enclosed chamber,
- Known specific location of target



Single particle detection
- High-Q Resonators
  - difficult coupling
  - Small interaction volume
- Digital PCR, Simoa



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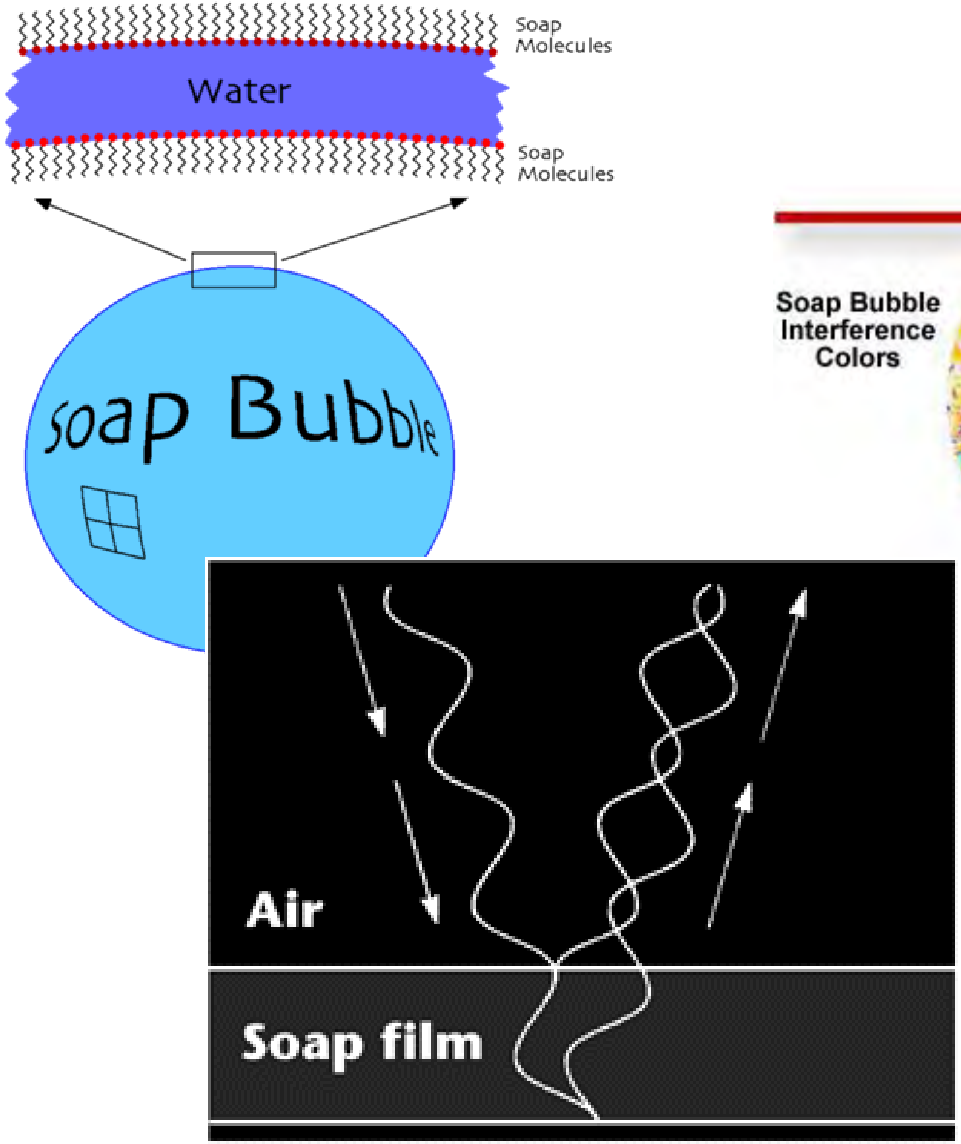
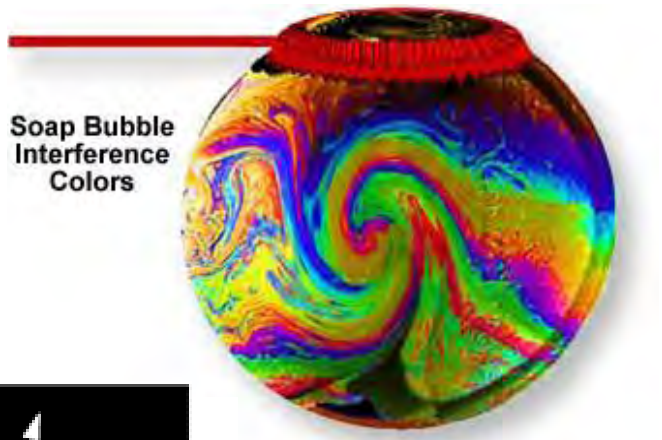
Optical Interference: from basic to the ultimate



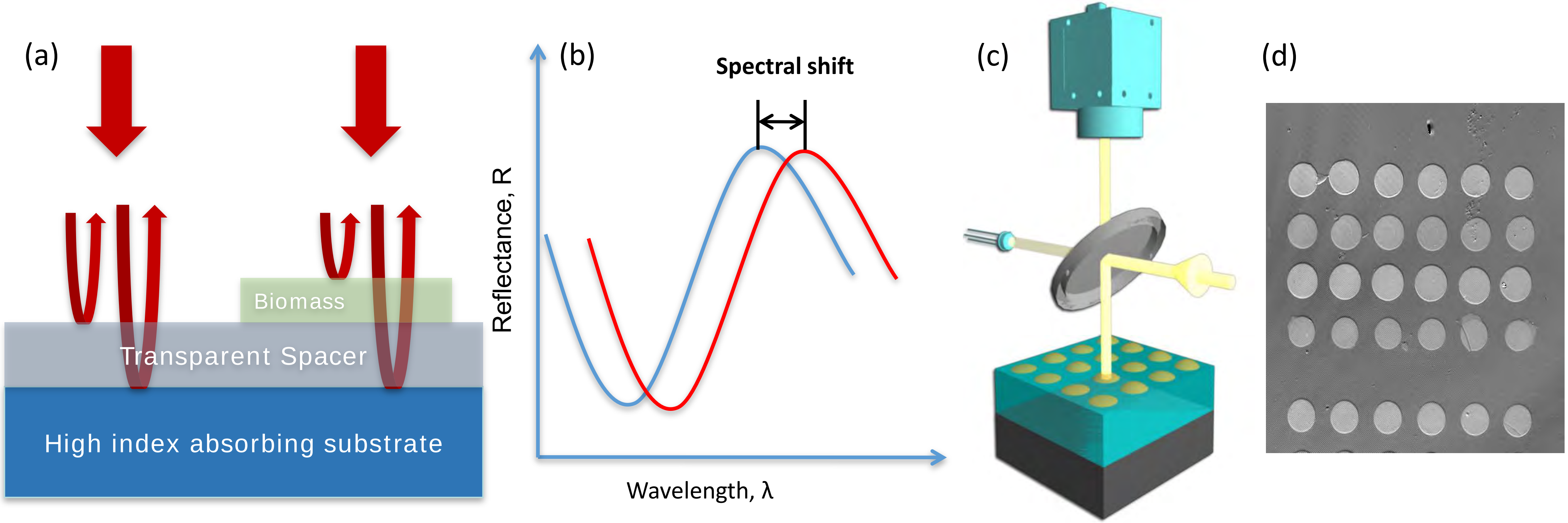


~ 10 nm thickness change can be visually observed

>10 orders of magnitude

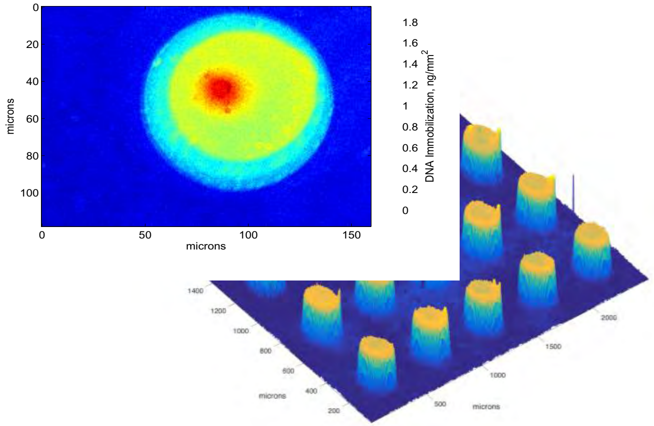
0.1 atto-m displacement

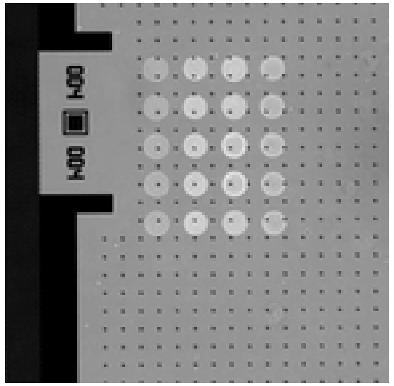
Interferometric Reflectance Imaging Sensor (**IRIS**)



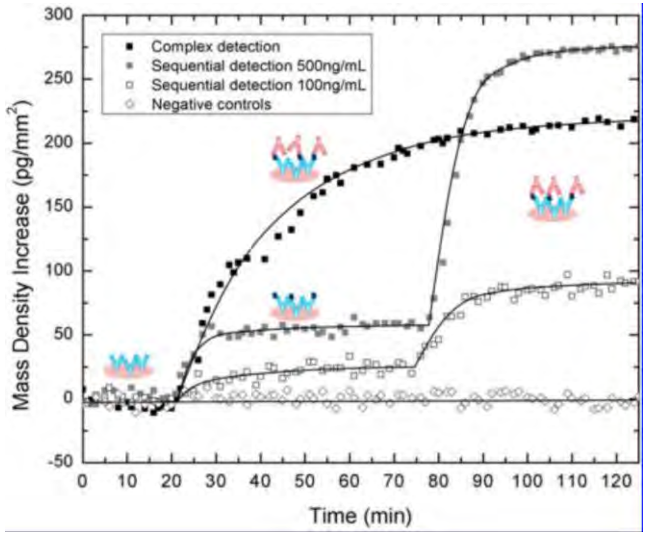
IRIS – a versatile biosensing technology platform for microarrays

Quantitative / QC

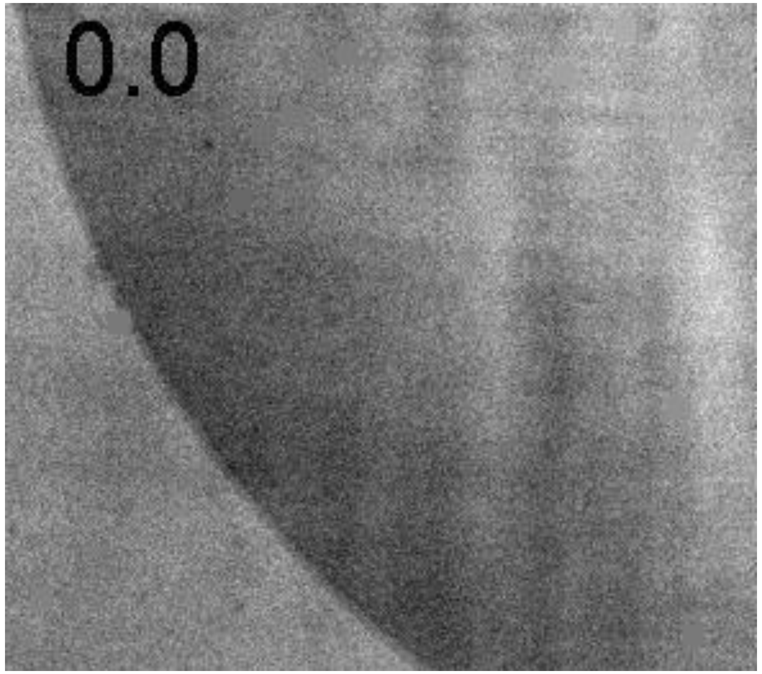




Dynamic Measurements



Single virus/exosome



- decade long R&D
- various applications demonstrated.
    - From QC to single exosome/virus detection
    - pg/mm^2 sensitivity
    - Single biological nanoparticle detection and characterization
    - attoM sensitivity for protein and nucleic acid detection

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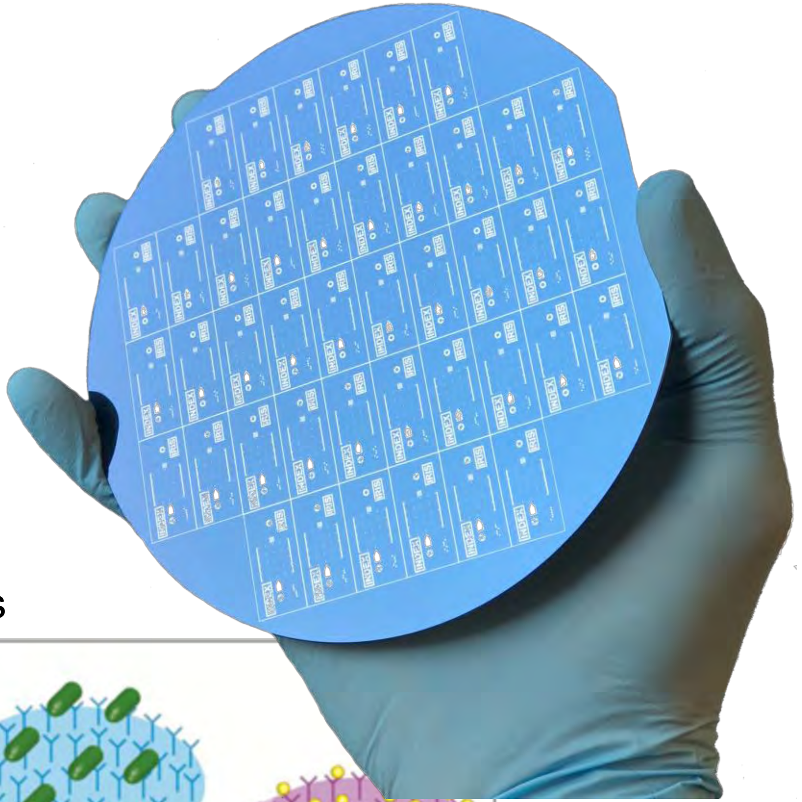
Sensor Chip Requirements

- Multilayer reflector with no stray light
- Flat and smooth surface
- Chemical functionalization / glass
- Manufacturable and scalable

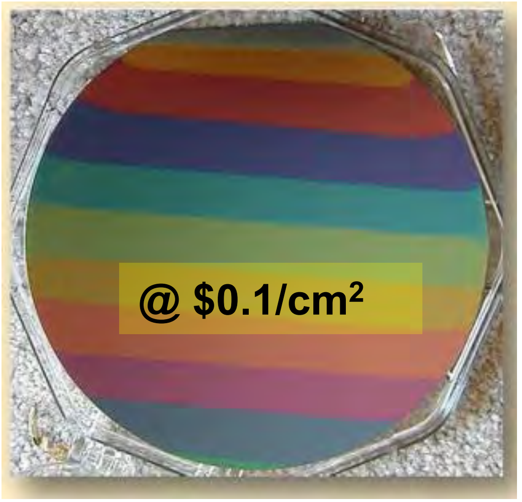
Transparent Spacer

High index absorbing substrate

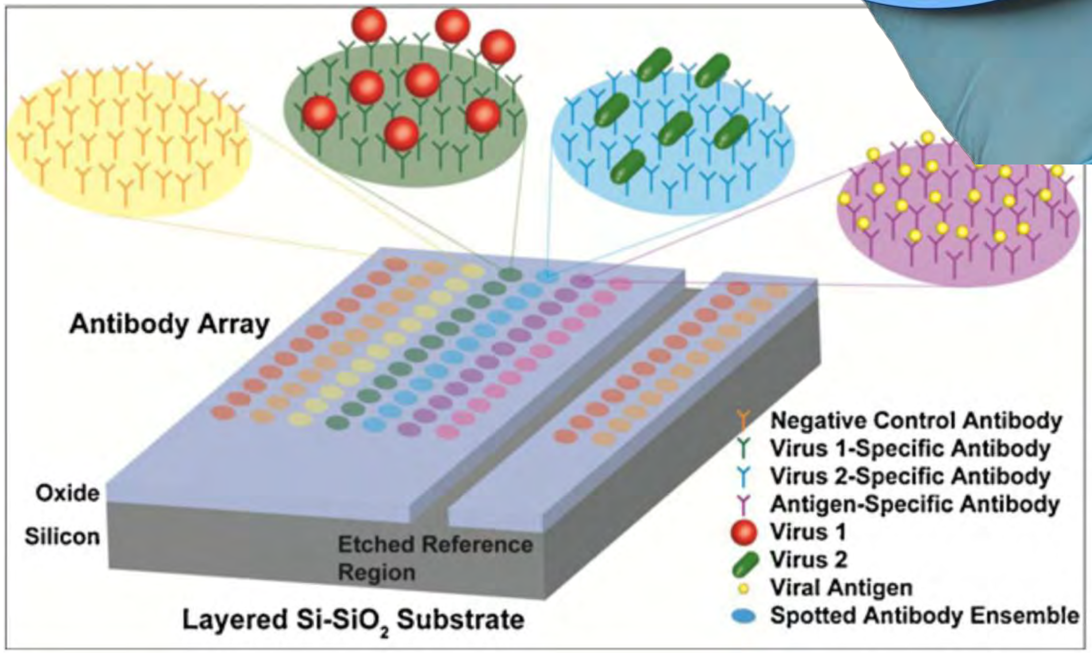


soap film

Oxide coated Si



Protein microarray chips with 100s to 1,000s of probe spots



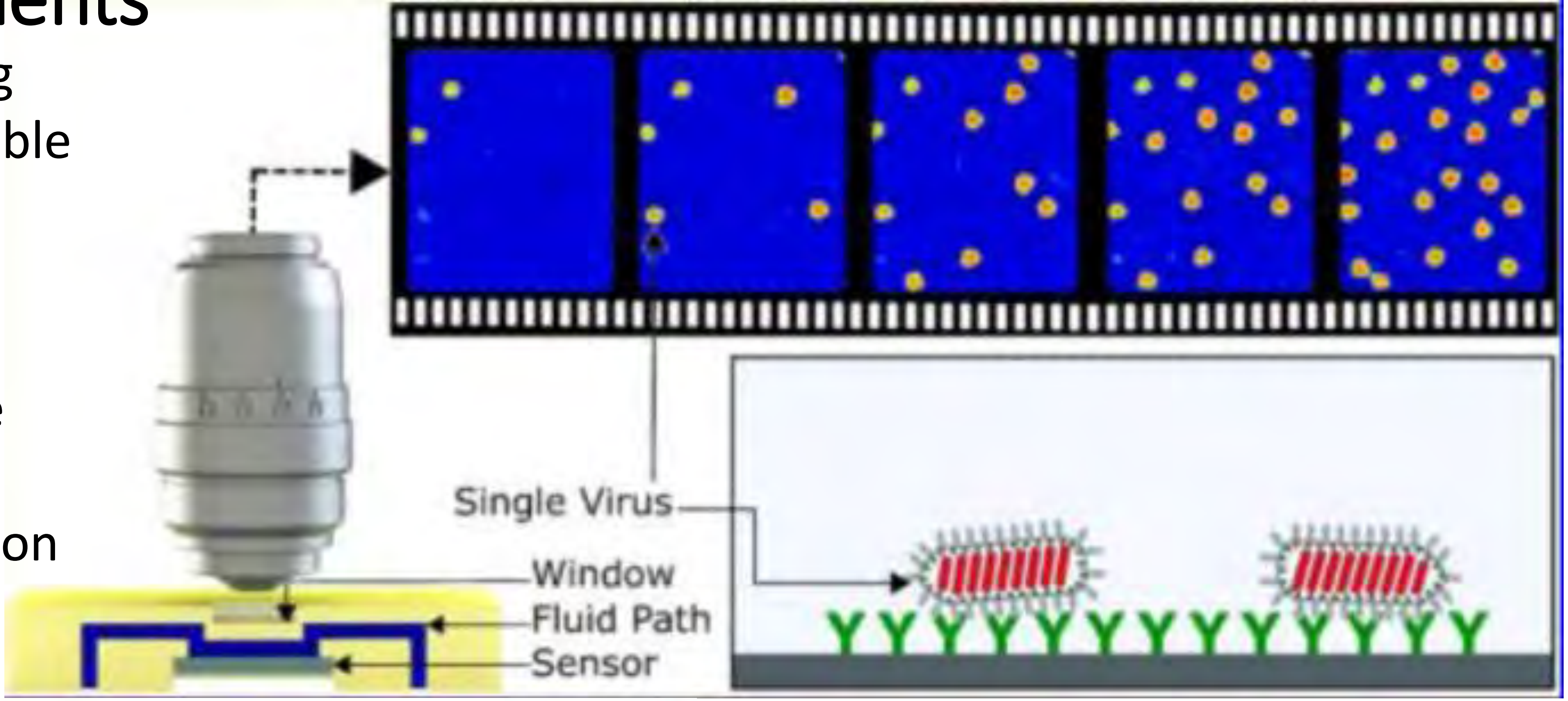
Ünlü et al, "STRUCTURED SUBSTRATES FOR OPTICAL SURFACE PROFILING,' US Patent No: 9599611, 2017

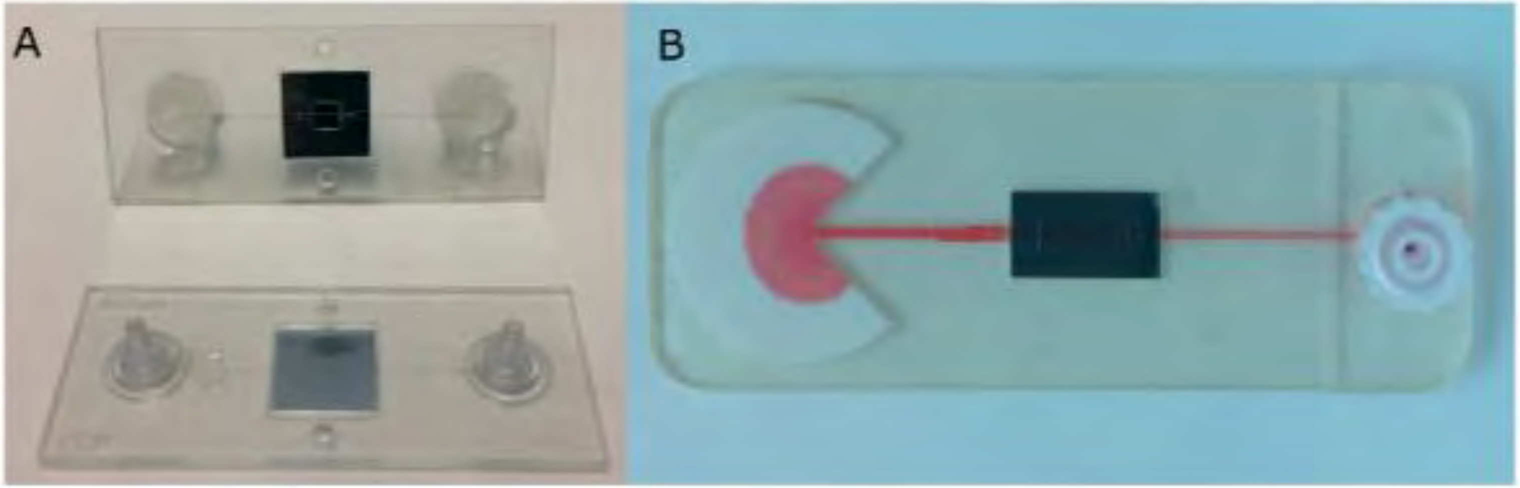
pg/mm^2 sensitivity 1,000s of spots

Cartridge Requirements

- Optical quality for imaging
- Manufacturable and scalable
- Easy assembly with chip
- Cost

- Multi-layer polycarbonate laminates (7 layers)
  - Good prototype solution
  - Cost remains high for medium volume

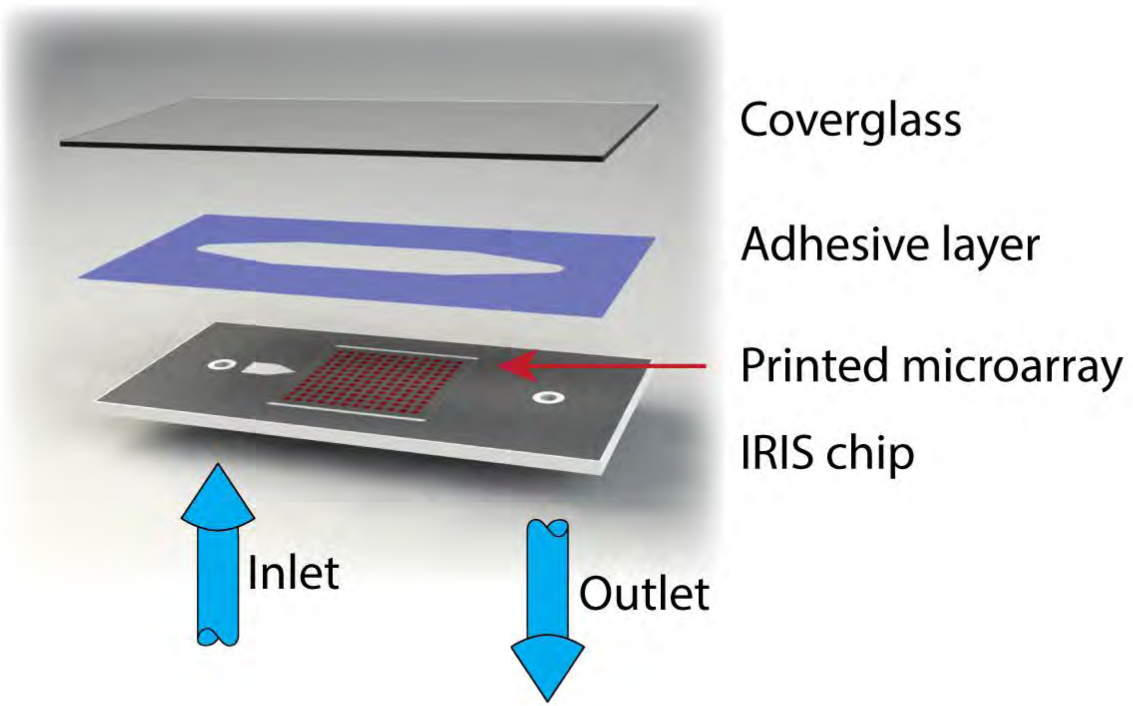


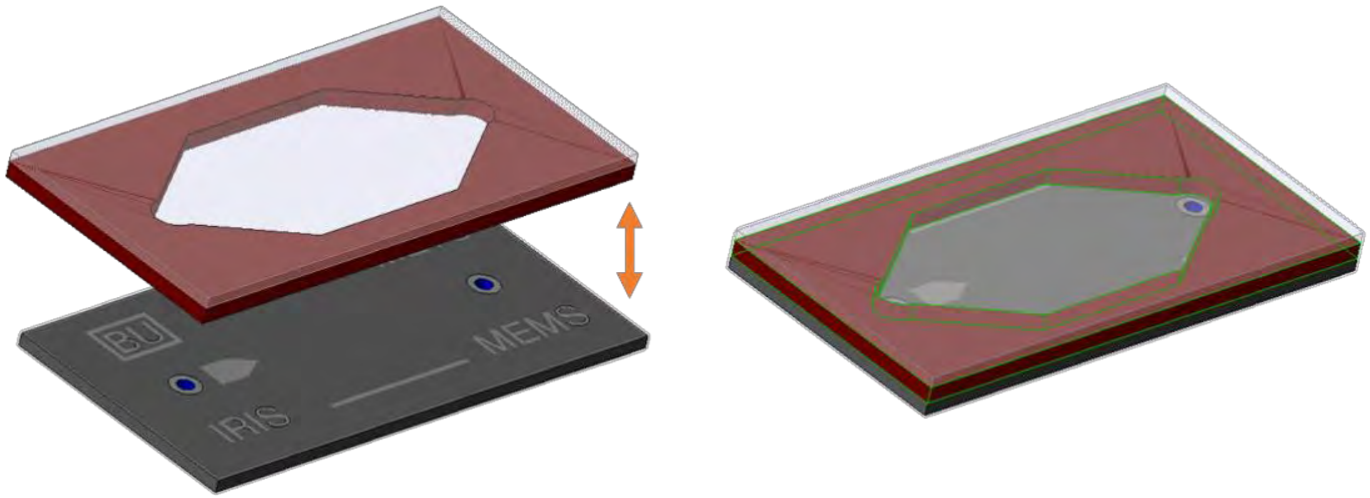


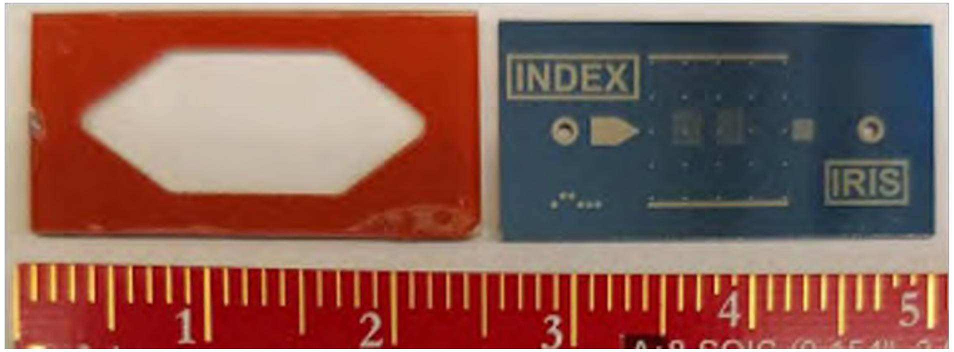
Scherr et al, *ACS Nano* 10 (2016)
Scherr et al, *Lab on a Chip* (2017)

Si-based Microfluidics solution

- Fluidics through Si
- Manufacturable / scalable – established infrastructure
- Top window can be separately optimized
  - AR coating
  - Polarization preserving

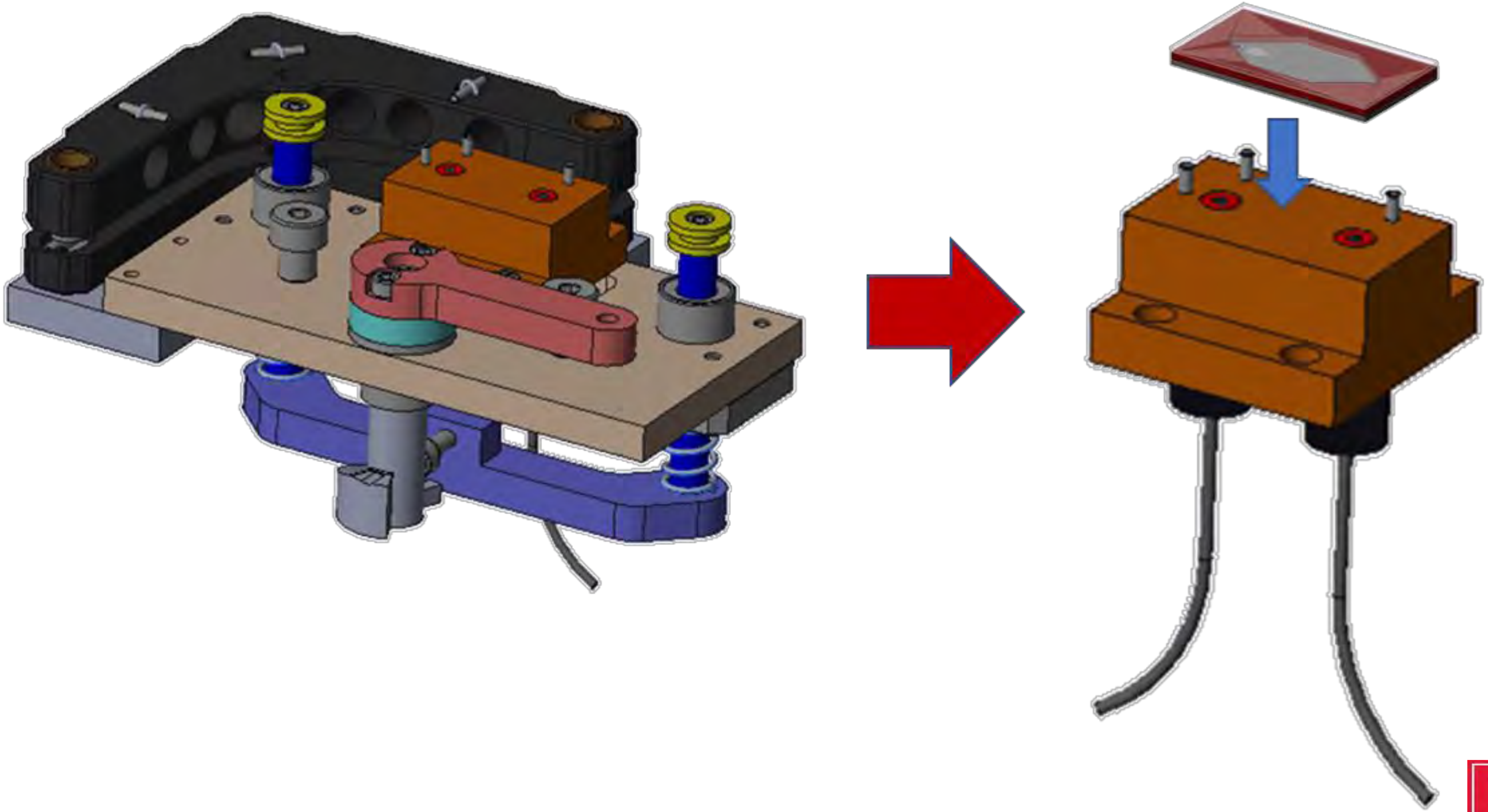






Yalcin-Ozkumur, JSTQE (2018)

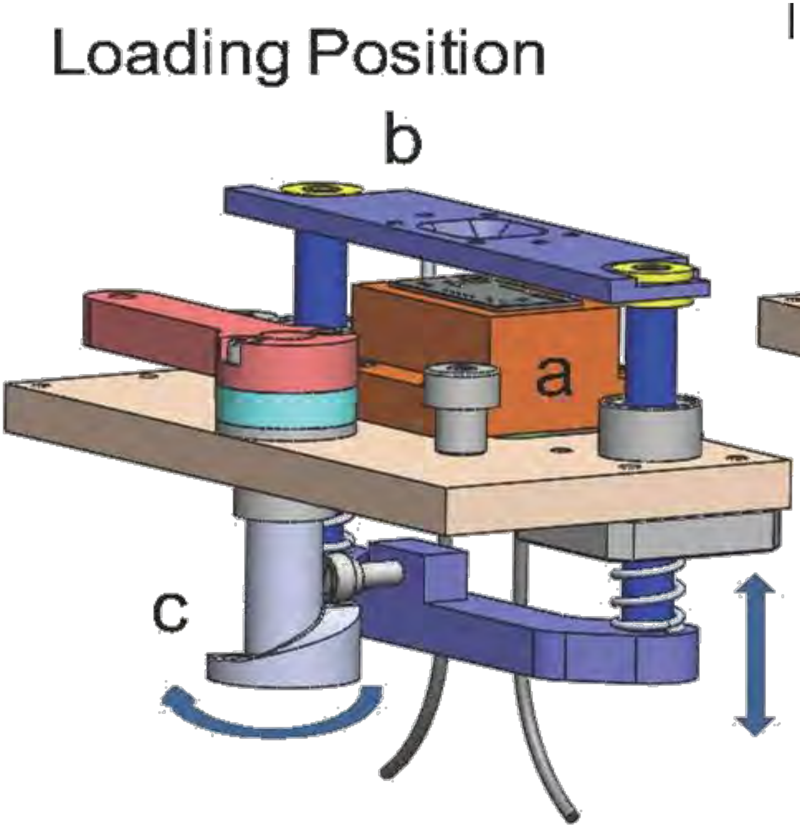
Mechanical Fixture

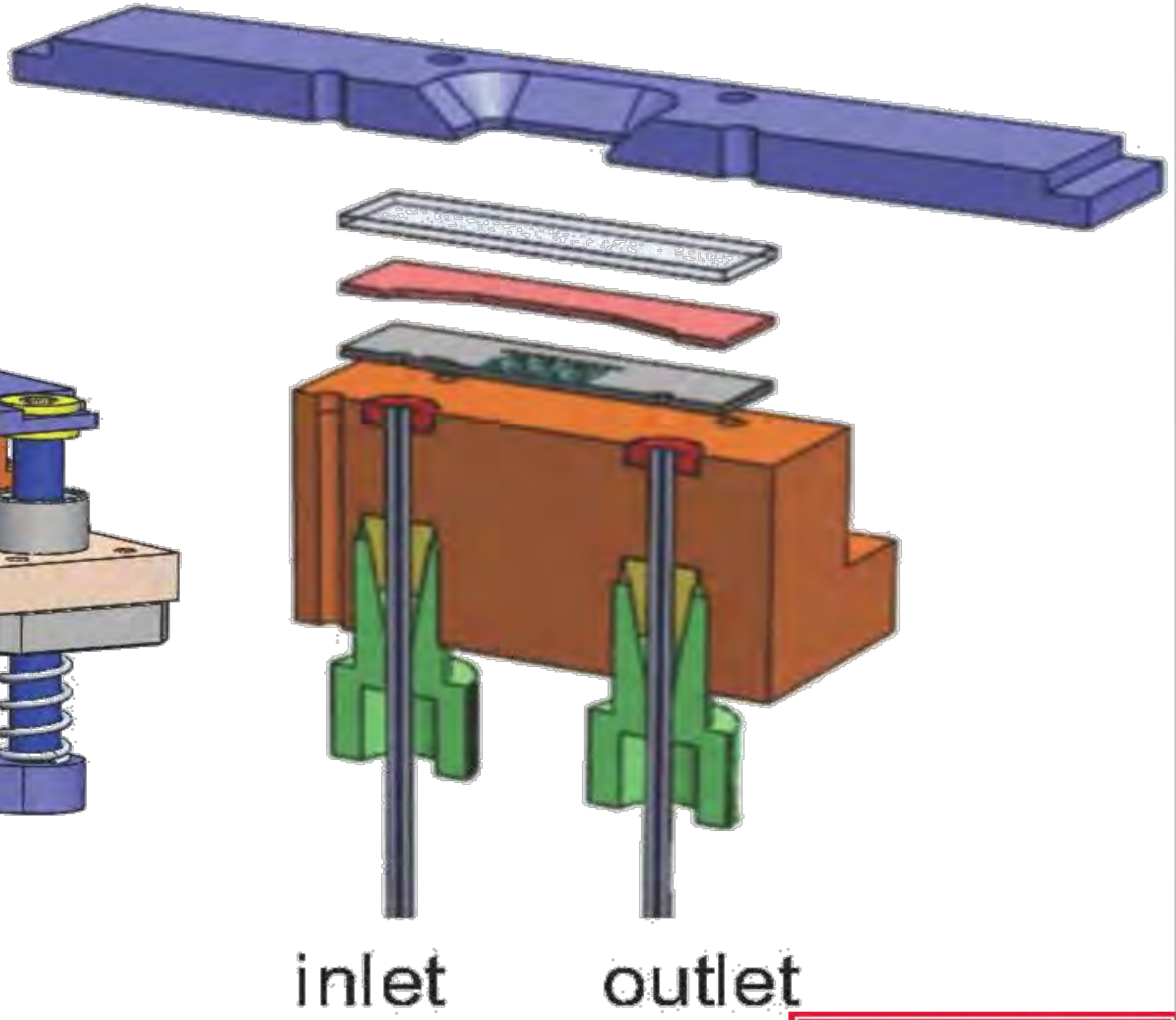


Cam Operation

Interferometric Detection and Enumeration of Viral Particles using Si-based Microfluidics

Ayca Yalcin Ozkumur, Fulya Ekiz Kanik, Jacob Trueb, Celalettin Yurdakul, and M. Selim Ünlü, *Fellow, IEEE*





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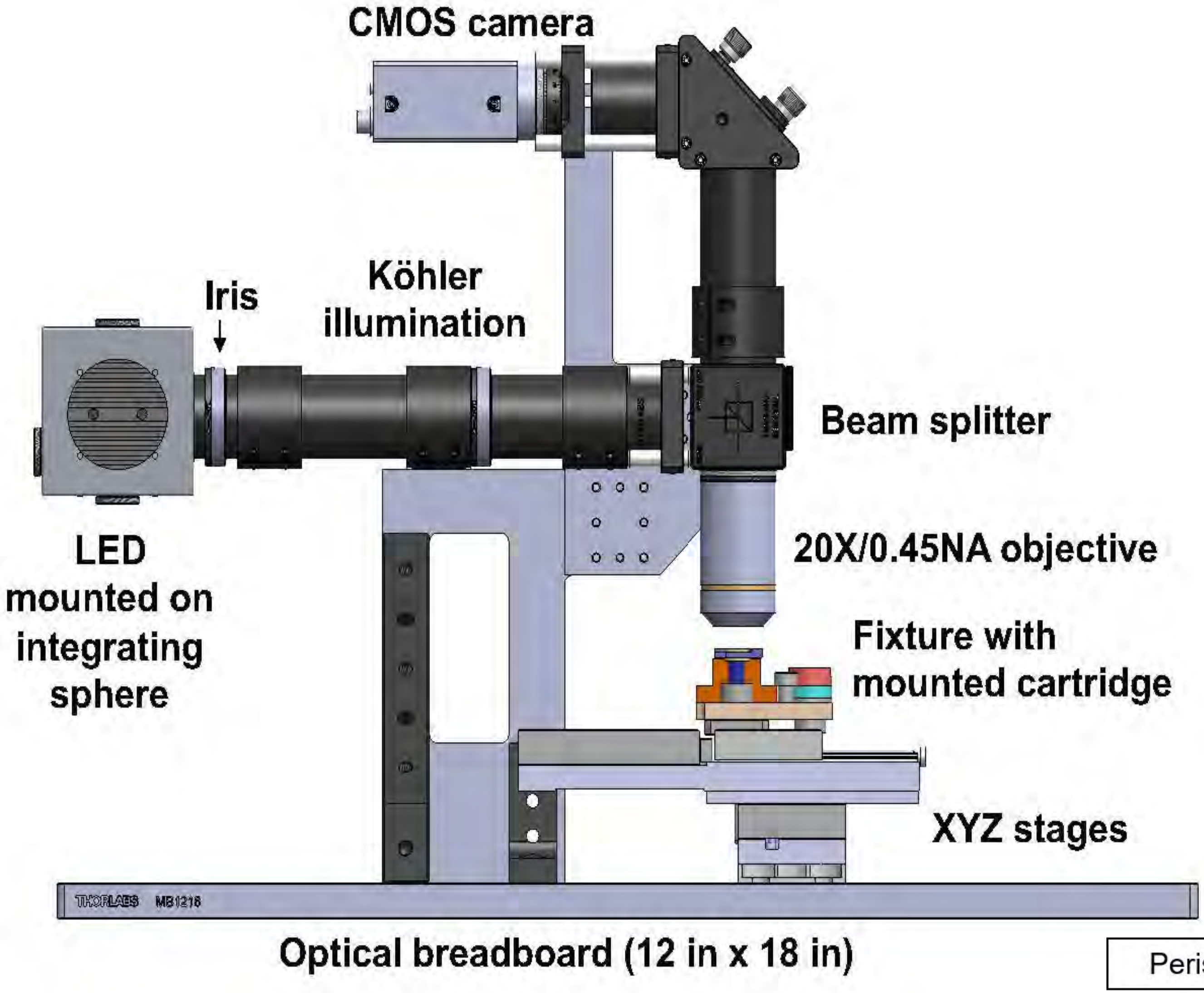
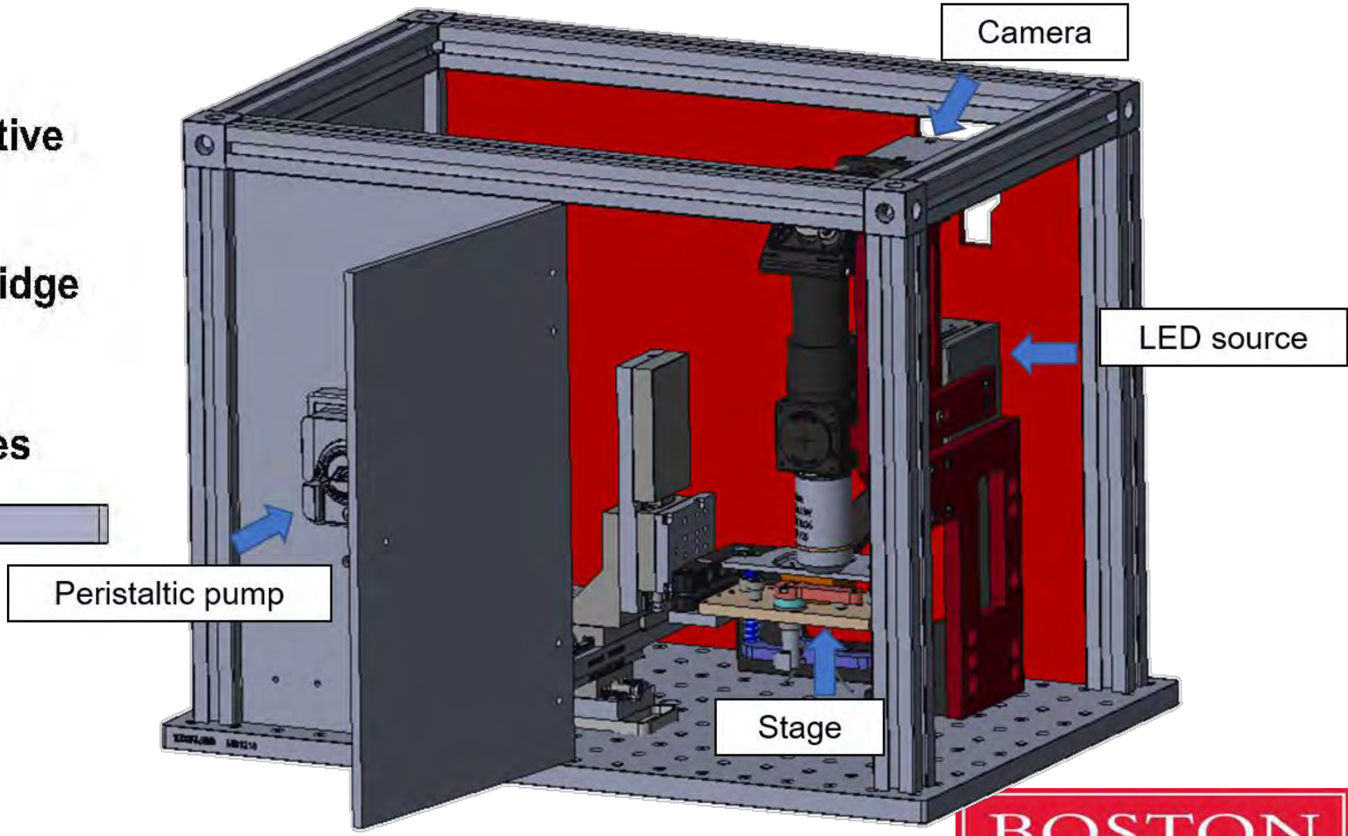
Prototype Instrument





Off-the-shelf components
Parts cost (BOM) under $10K

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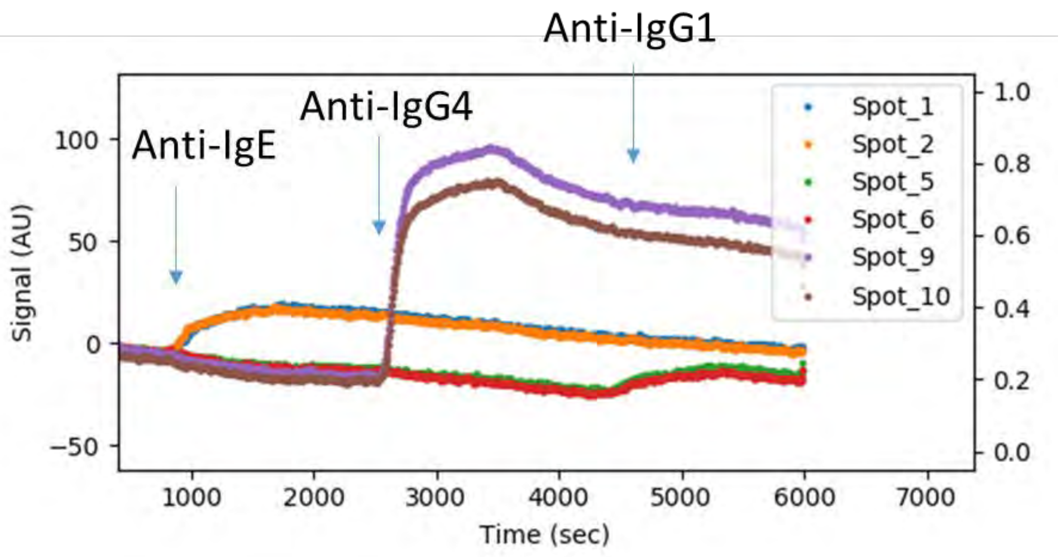
Outline – What can IRIS do?

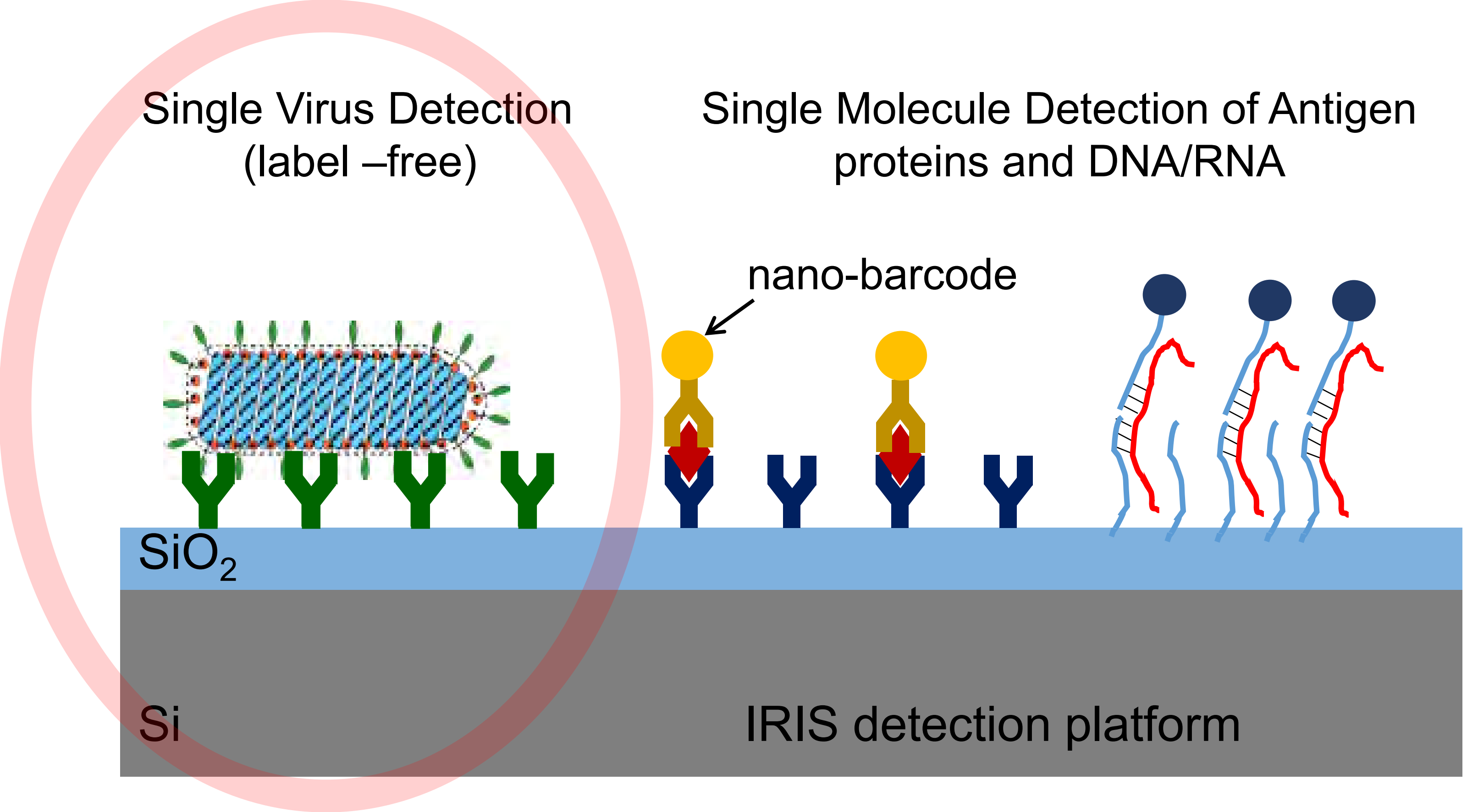
- Motivation
- Interferometric Reflectance **Imaging** Sensor
- Basic principles
- Sensor Chip and Cartridge Development
- Versatile platform
  1. Molecular kinetics
  2. Biological Nanoparticle Detection and Sizing
  3. Resolution improvement by oblique illumination and reconstruction
  4. Digital Microarrays (protein, DNA, RNA, miRNA)
- Conclusions and Future

1. Molecular binding Kinetic Measurements



- Multiplexed (100s to 1000s of probes)
- Quantitative
- Glass surface
- Inexpensive instrumentation and disposables
- ng/ml target sensitivity
- ~kDa molecular weight

2. Single Particle (Digital) Detection



- Label Free direct sensing of individual viruses
- Digital Detection: Single molecule level detection of Nucleic Acids and Proteins

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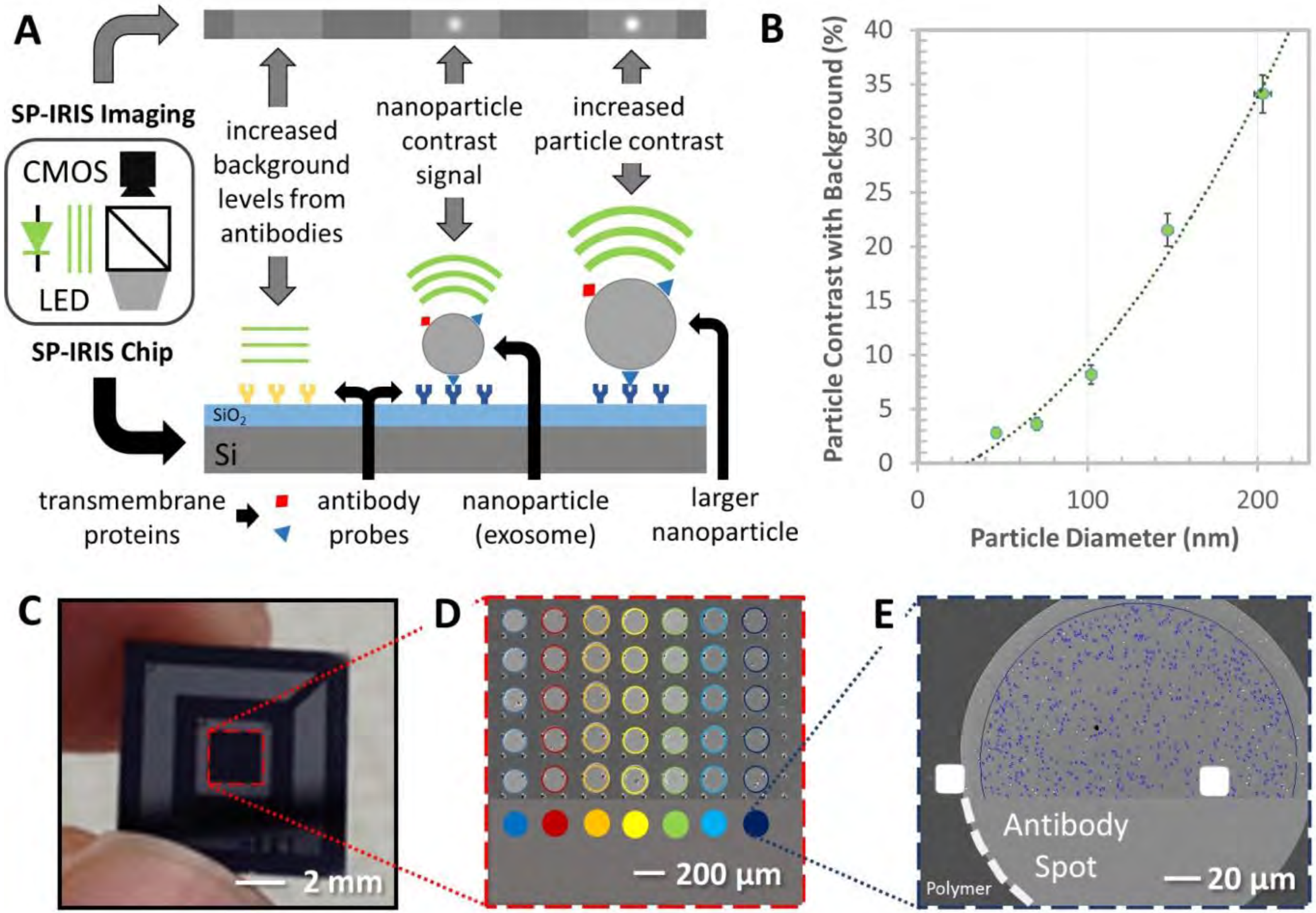
Simple Particle Detection



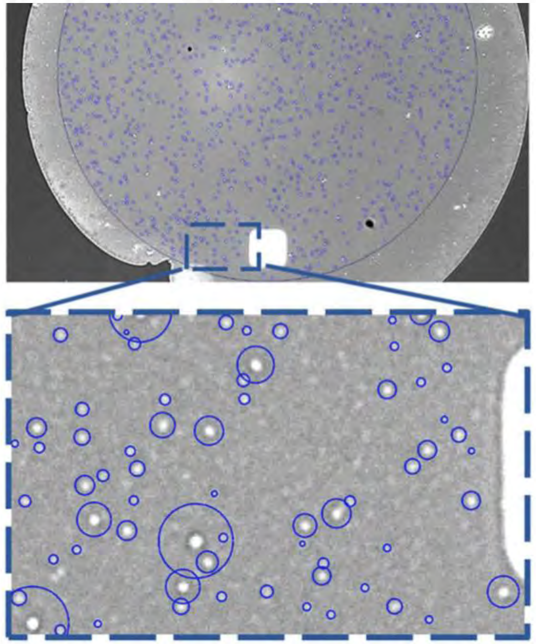
SCIENTIFIC REPORTS

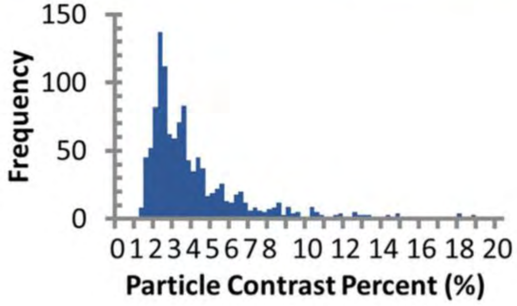
OPEN

Digital Detection of Exosomes by Interferometric Imaging

George G. Daaboul[1,*], Paola Gagni[2,*], Luisa Benussi[3], Paolo Bettotti[4], Miriam Ciani[3], Marina Cretich[2], David S. Freedman[1], Roberta Ghidoni[3], Ayca Yalcin Ozkumur[5], Chiara Piotto[4], Davide Prosperi[6], Benedetta Santini[6], M. Selim Ünlü[7] & Marcella Chiari[2]

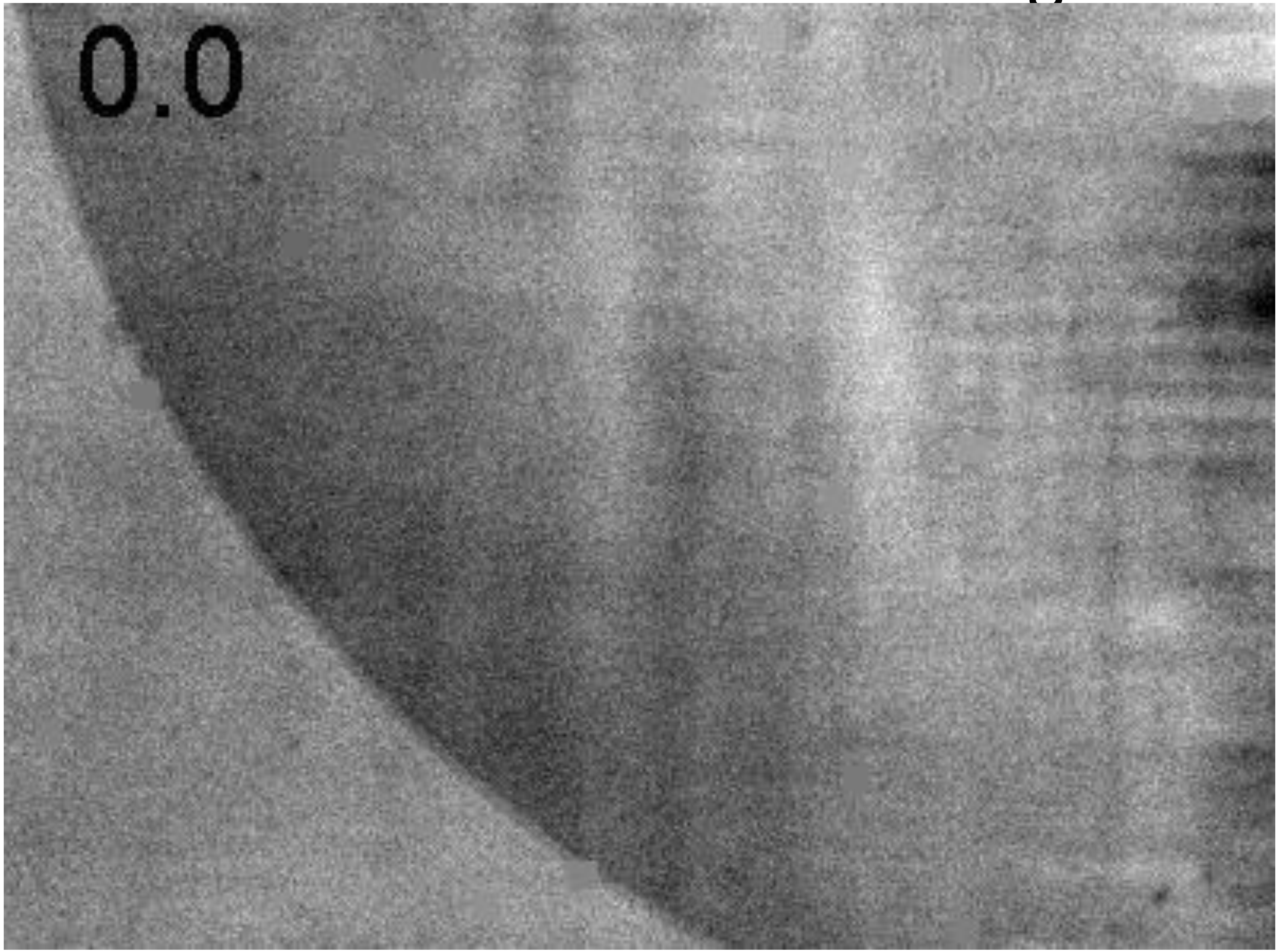
2016

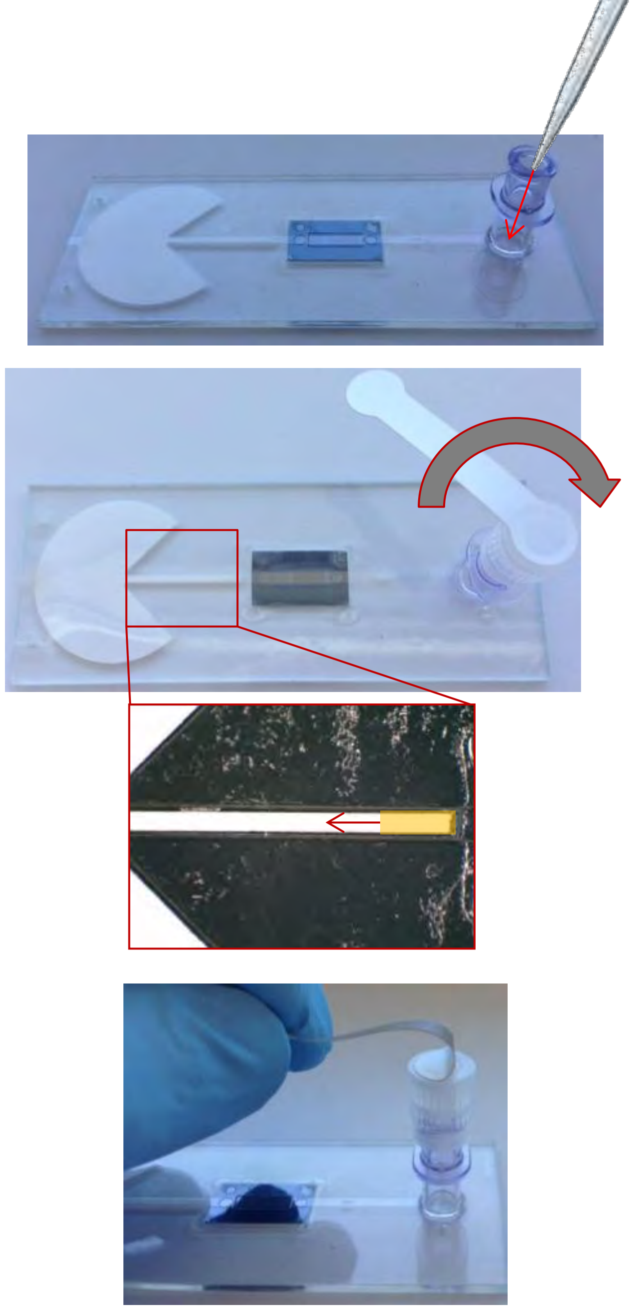




Virus Detection in Passive Cartridge

1.
2.
3.
4.
5.





Lab on a Chip

PAPER

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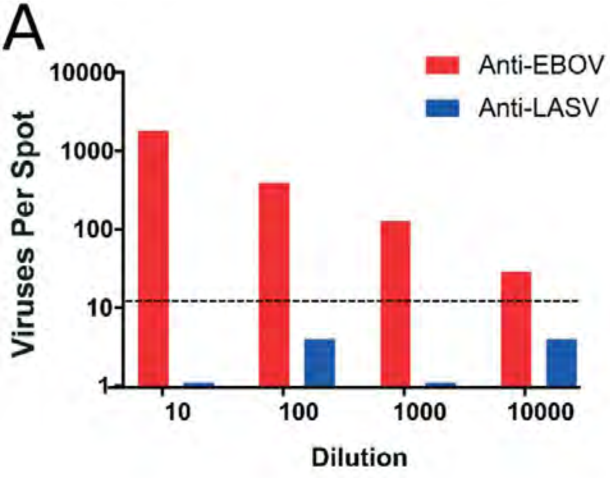
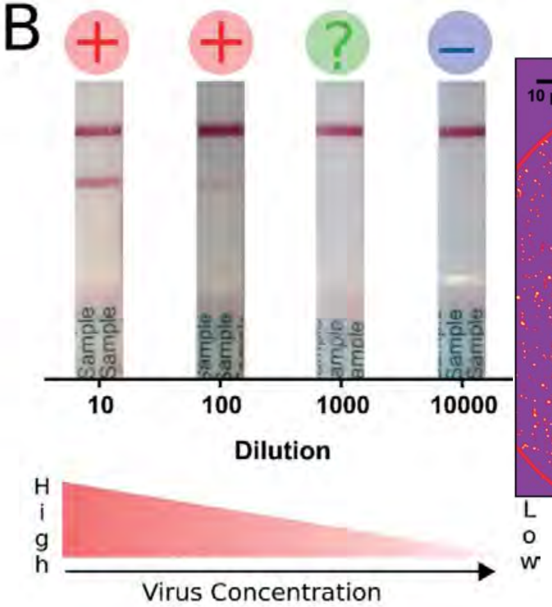
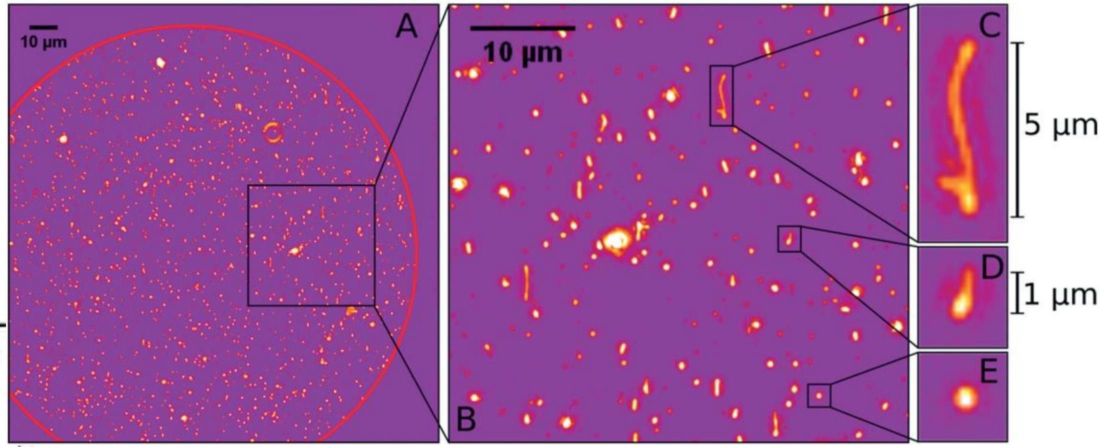
Cite this: DOI: 10.1039/c6lc01528j

Disposable cartridge platform for rapid detection of viral hemorrhagic fever viruses†

Steven M. Scherr,[a] David S. Freedman,[b] Krystle N. Agans,[cd] Alexandru Rosca,[b] Erik Carter,[e] Melody Kuroda,[f] Helen E. Fawcett,[a] Chad E. Mire,[cd] Thomas W. Geisbert,[cd] M. Selim Ünlü[ghi] and John H. Connor*[eh]







Dilution	SP-IRIS Anti-EBOV (Viruses Per Spot)	SP-IRIS Anti-LASV (Viruses Per Spot)	ReEbov Test Strip
1:10	1824	**	+
1:100	393	4	+
1:1000	126	-4	?
1:10000	29	4	-

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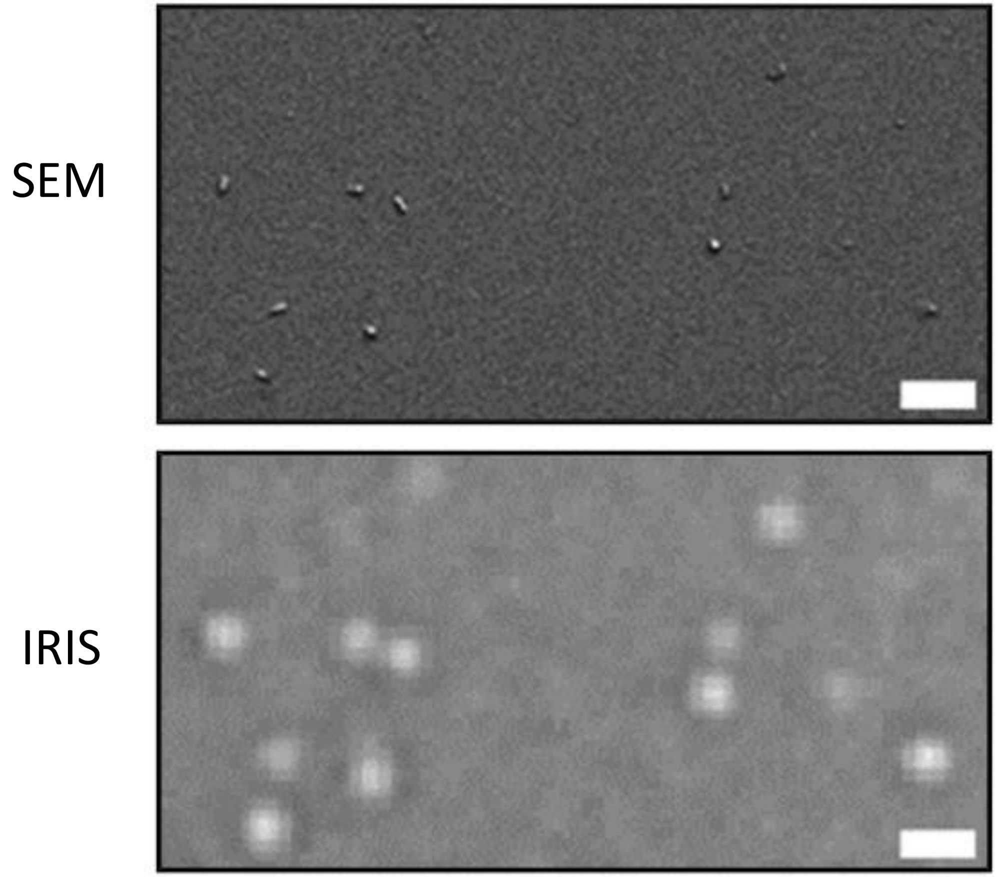
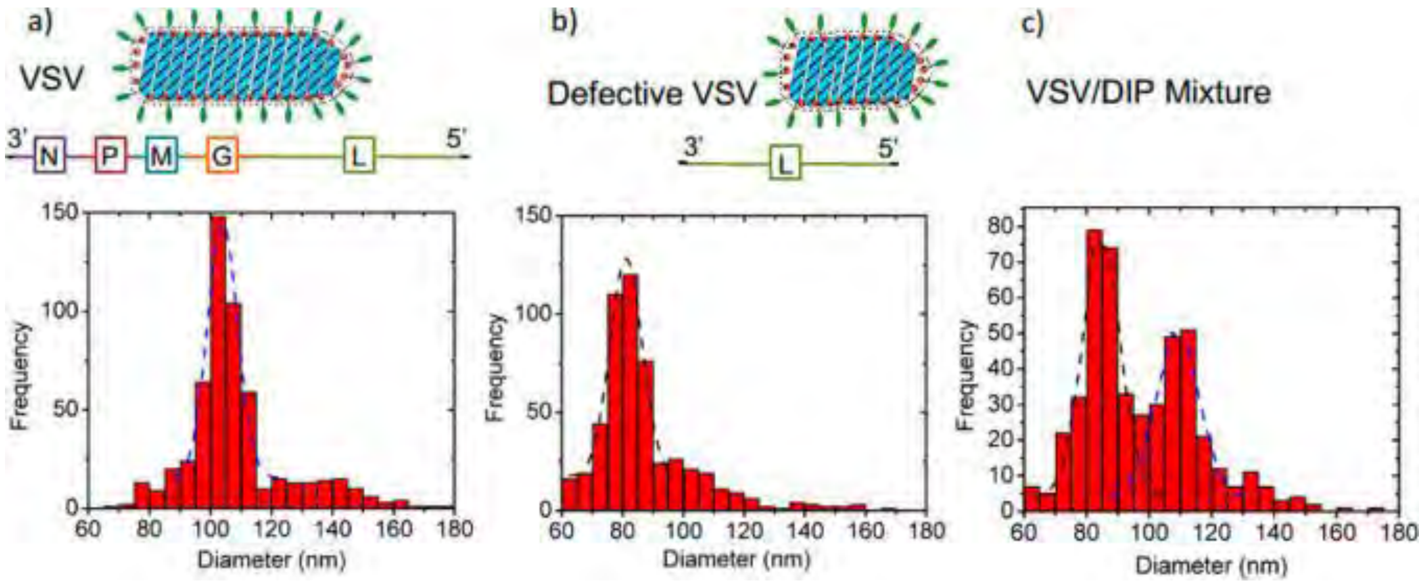
2b. Virus characterization: size determination





Daaboul et al, *ACS nano* 8 (2014)

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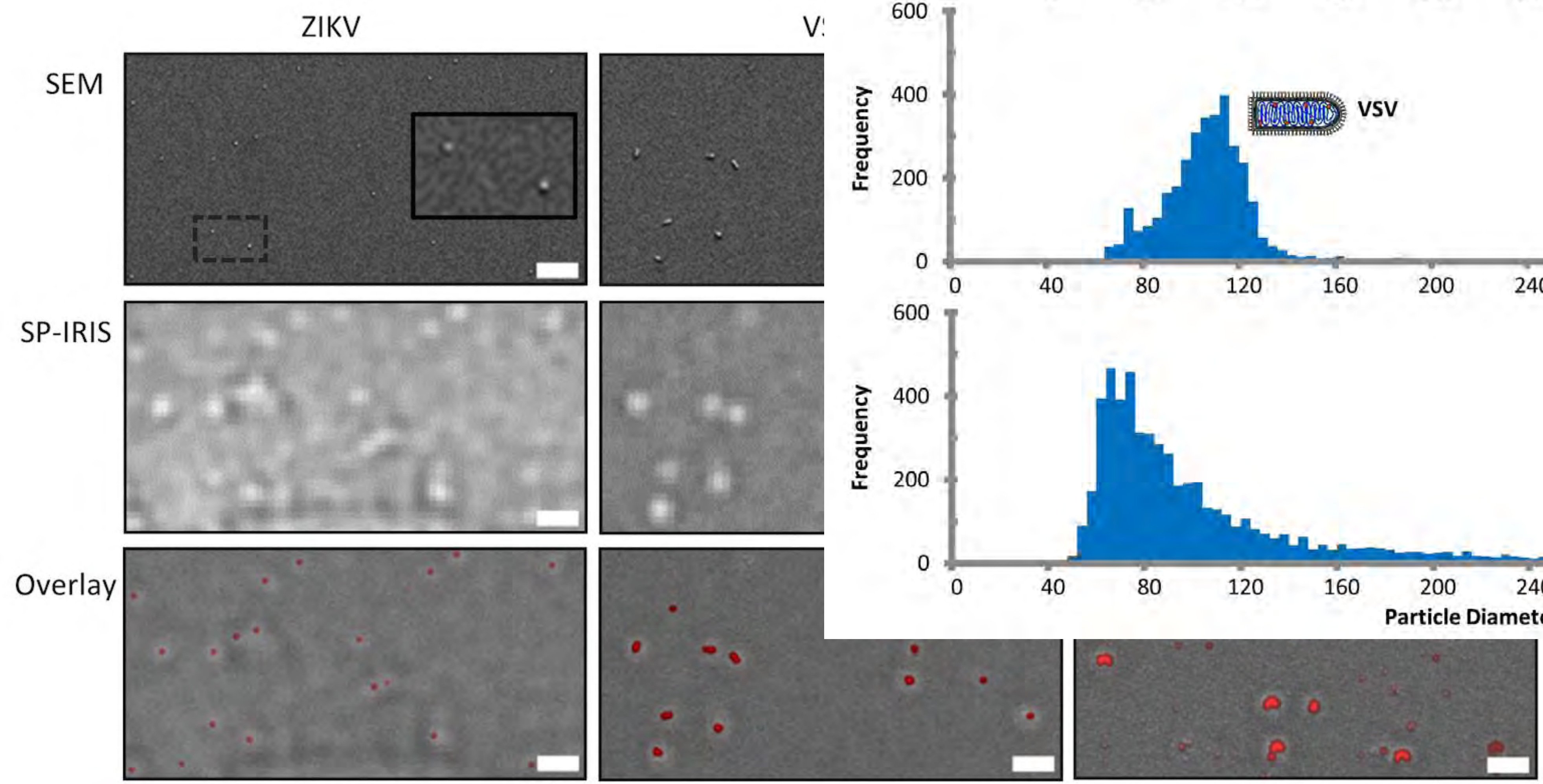
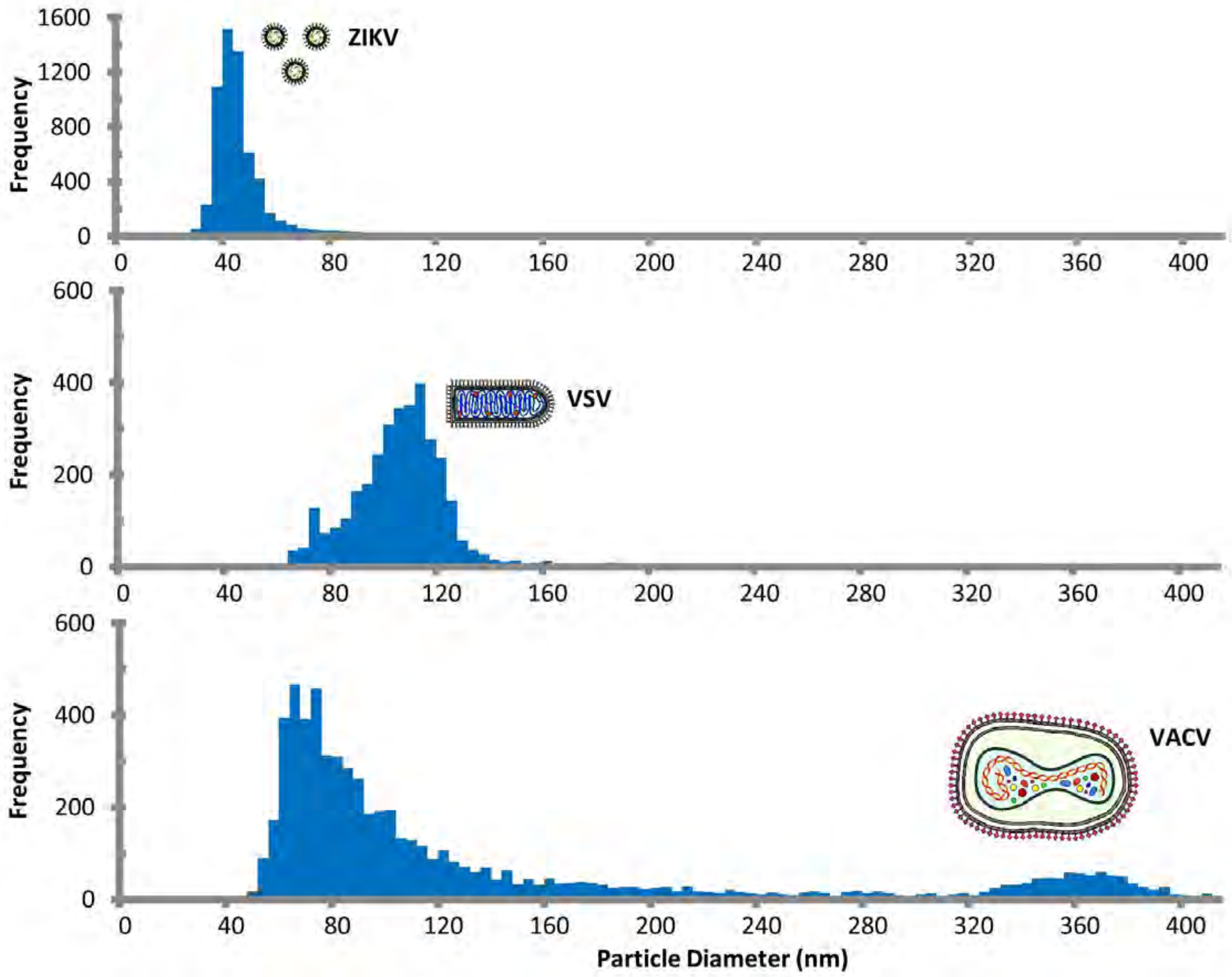
Various viruses

GG Daaboul, et al. PloS one (2017)





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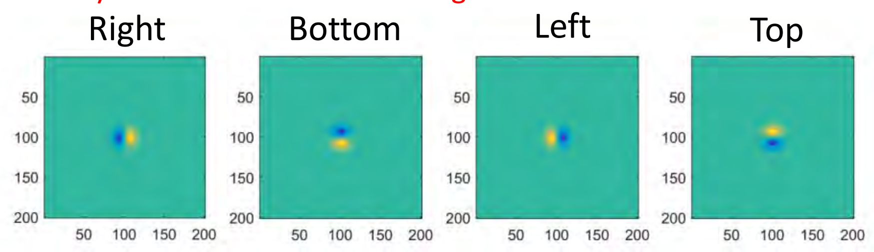
3. Super-resolution in wide-field interferometric microscopy

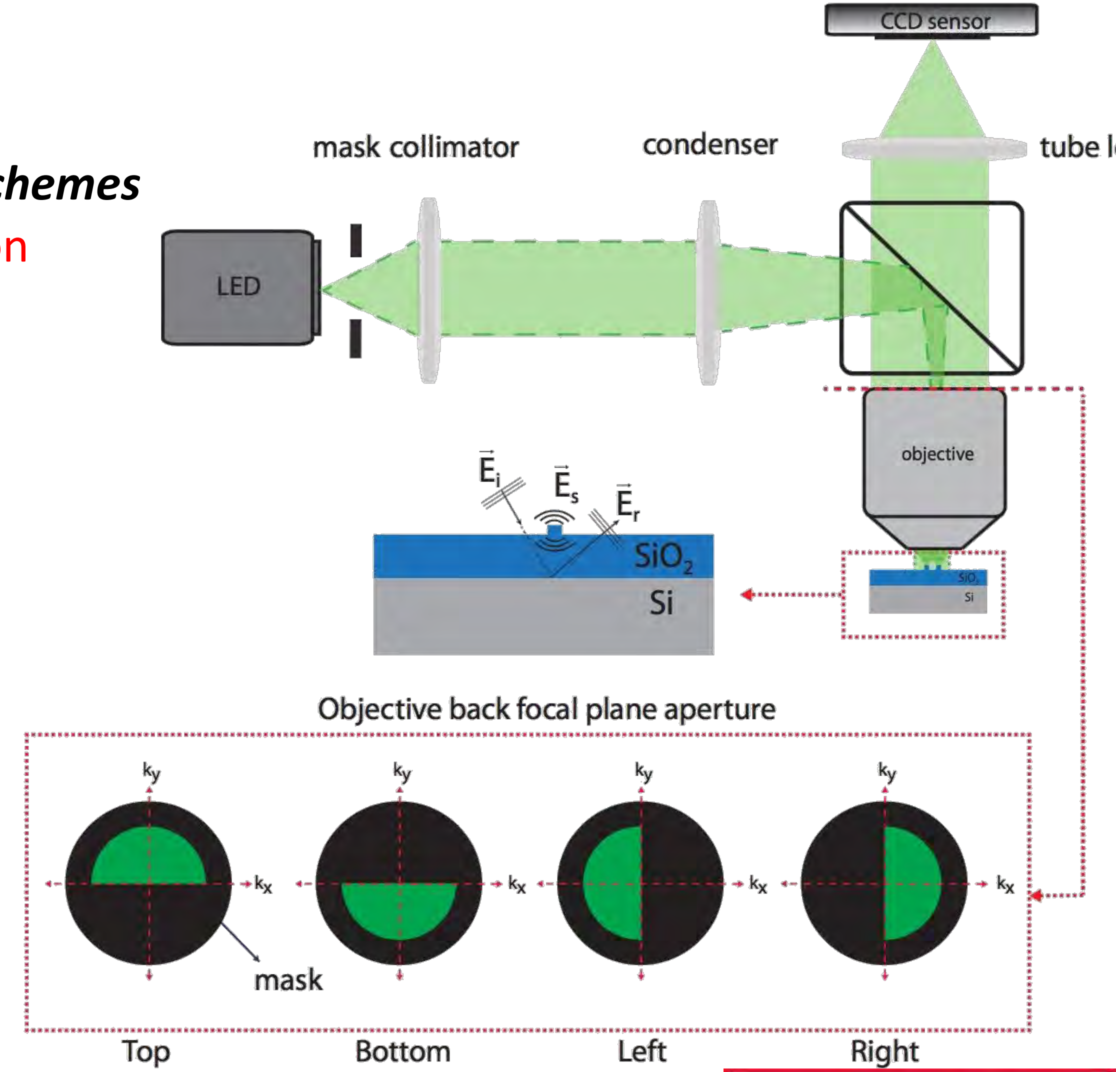
- ***Enhancing low-index nanoparticle resolution via reconstruction schemes***
  Asymmetric illumination based reconstruction for super resolution
  (with Lei Tian)

$$\min_x \sum_{j=1}^{N} \|A_j x - y_j\|_2^2 + \alpha \|x\|_2^2$$

$$x = \mathcal{F}^{-1}\left\{ \frac{\sum_{j=1}^{N} H_j^* y_j}{\sum_{j=1}^{N} |H_j|^2 + \alpha} \right\}$$

Fourier transforms of the transfer functions (H) for each asymmetric illumination configuration





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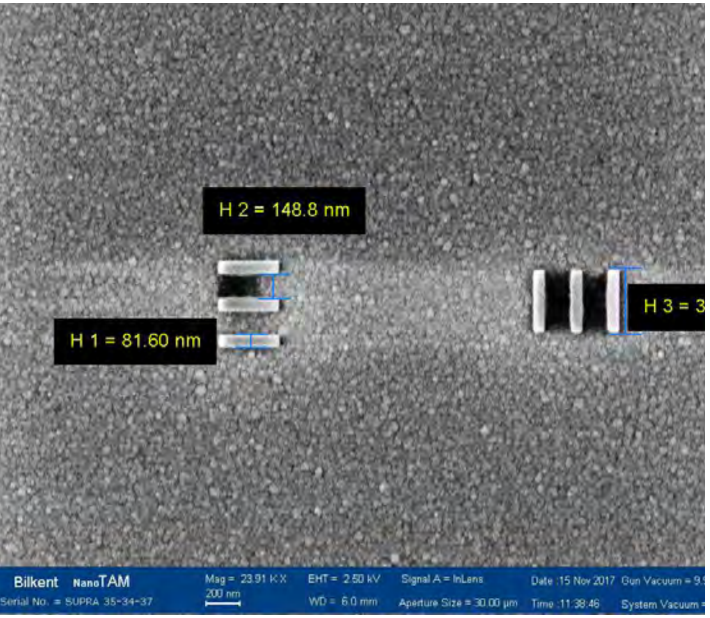
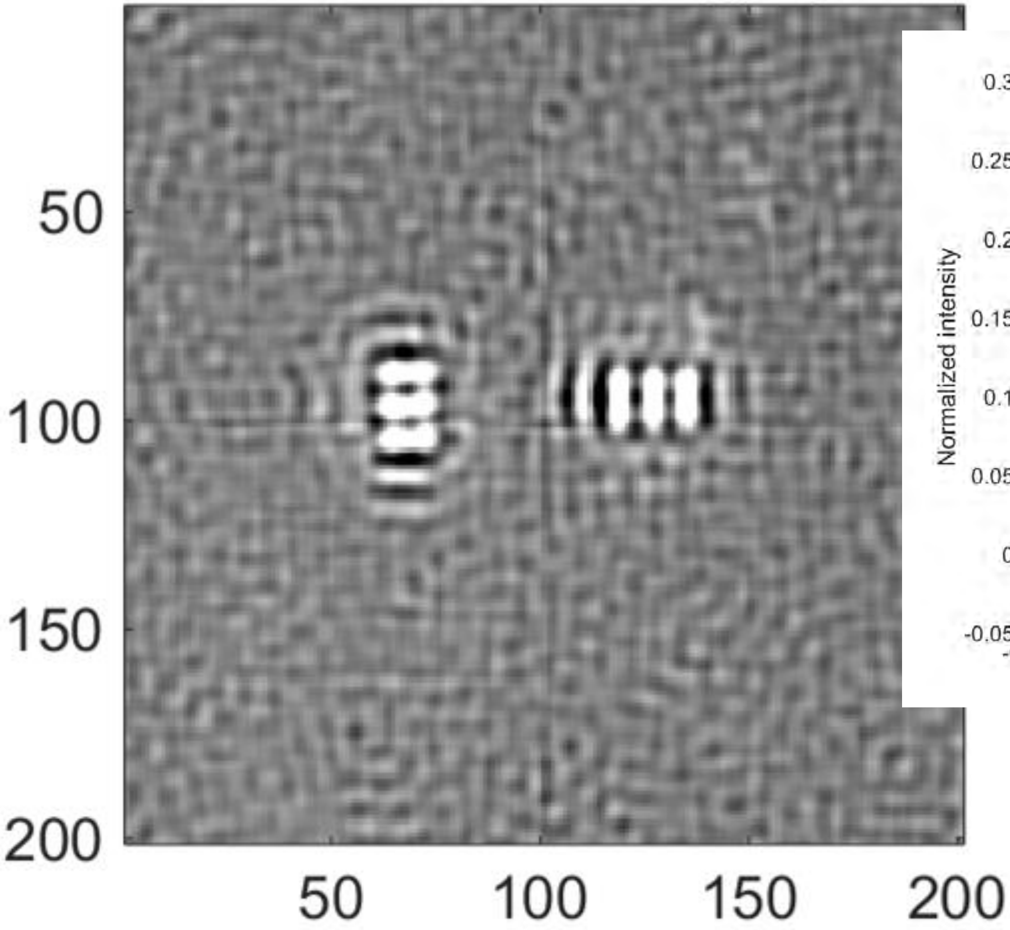
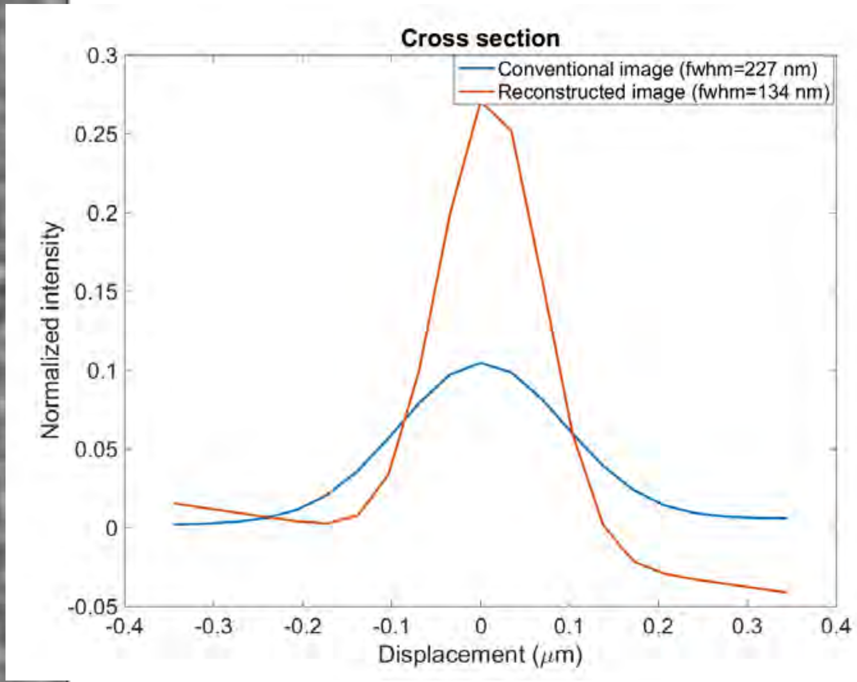
150 nm separation, 0.9 NA, λ=420nm







FWHM ~ 130nm < (λ / 3)

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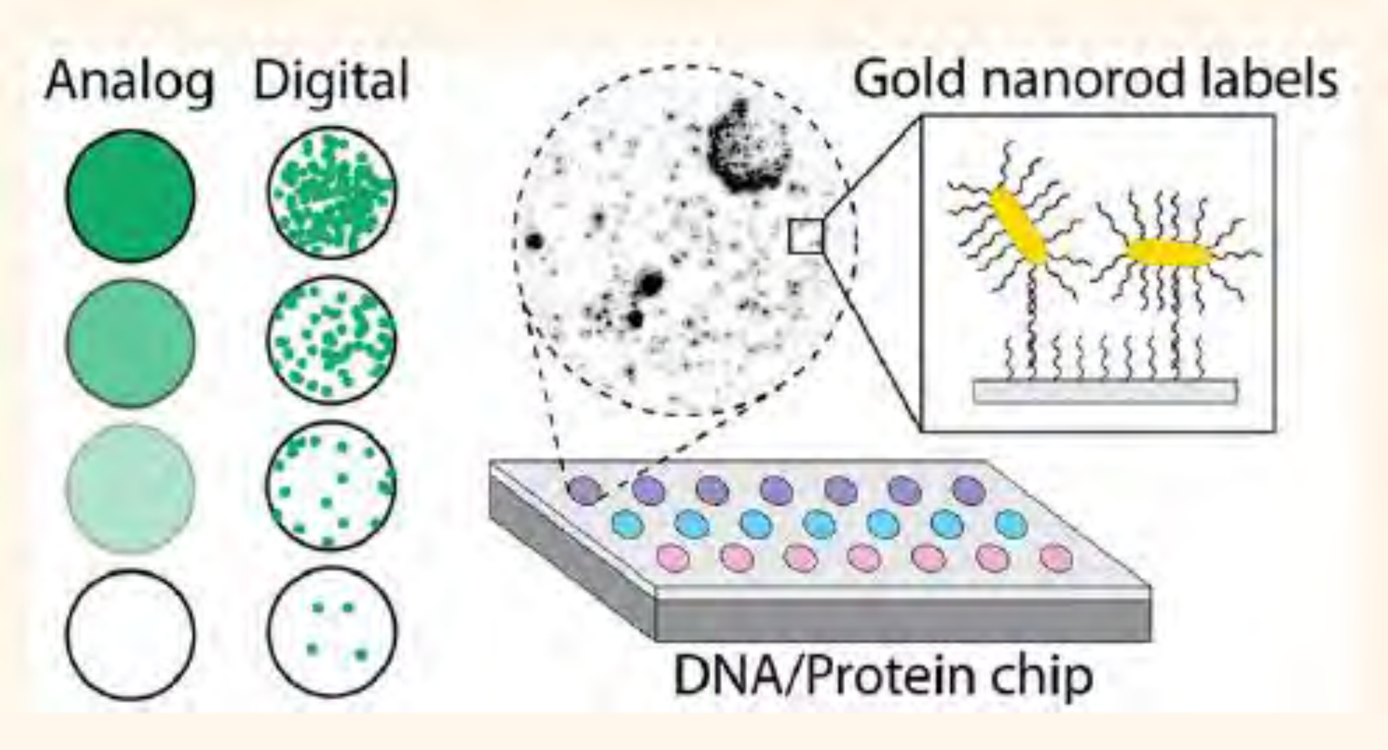
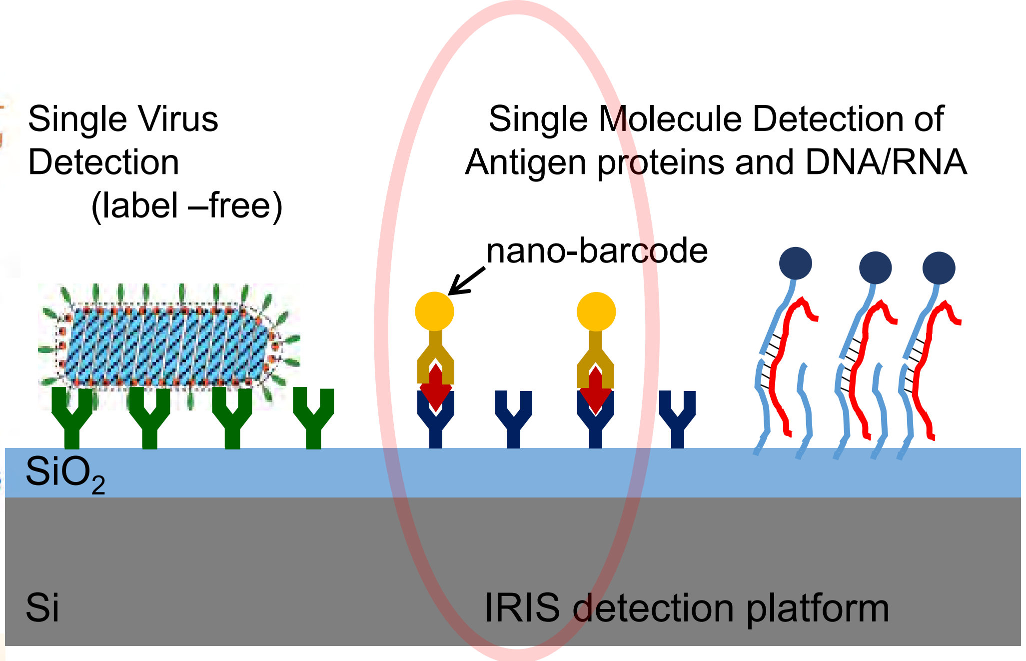
ACS NANO
Cite This: ACS Nano XXXX, XXX, XXX−XXX
www.acsnano.org

4. Digital Microarrays: Single-Molecule Readout with Interferometric Detection of Plasmonic Nanorod Labels

Derin Sevenler,*[,†] George G. Daaboul,[‡] Fulya Ekiz Kanik,[†] Neşe Lortlar Ünlü,[§] and M. Selim Ünlü[†,§]

[†]Department of Electrical and Computer Engineering and [§]Department of Biomedical Engineering, Boston University, Boston, Massachusetts 02215, United States
[‡]NanoView Biosciences, Boston, Massachusetts 02215, United States





- Digital means counting
- 10,000-fold more sensitive than commercial microarrays, while maintaining all of the advantages:
  1. Highly multiplexed
  2. Low cost
  3. Fast
- Application: molecular diagnostics

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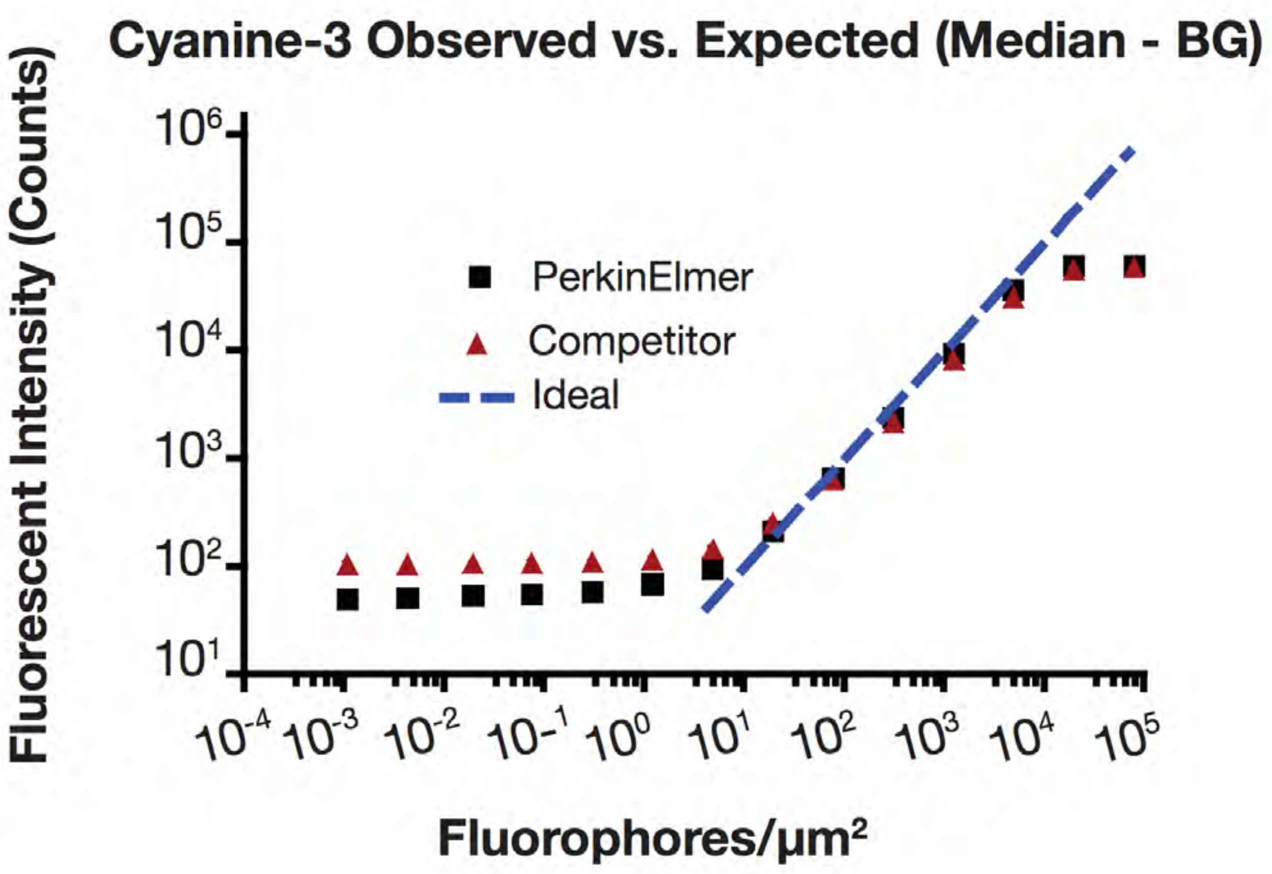
Performance limits of fluorescence

- Sensitivity limit: 10 fluorophores/um^2
- Dynamic range: 100-1000

… but microarray spots are 10,000 um^2

~100 μm diameter



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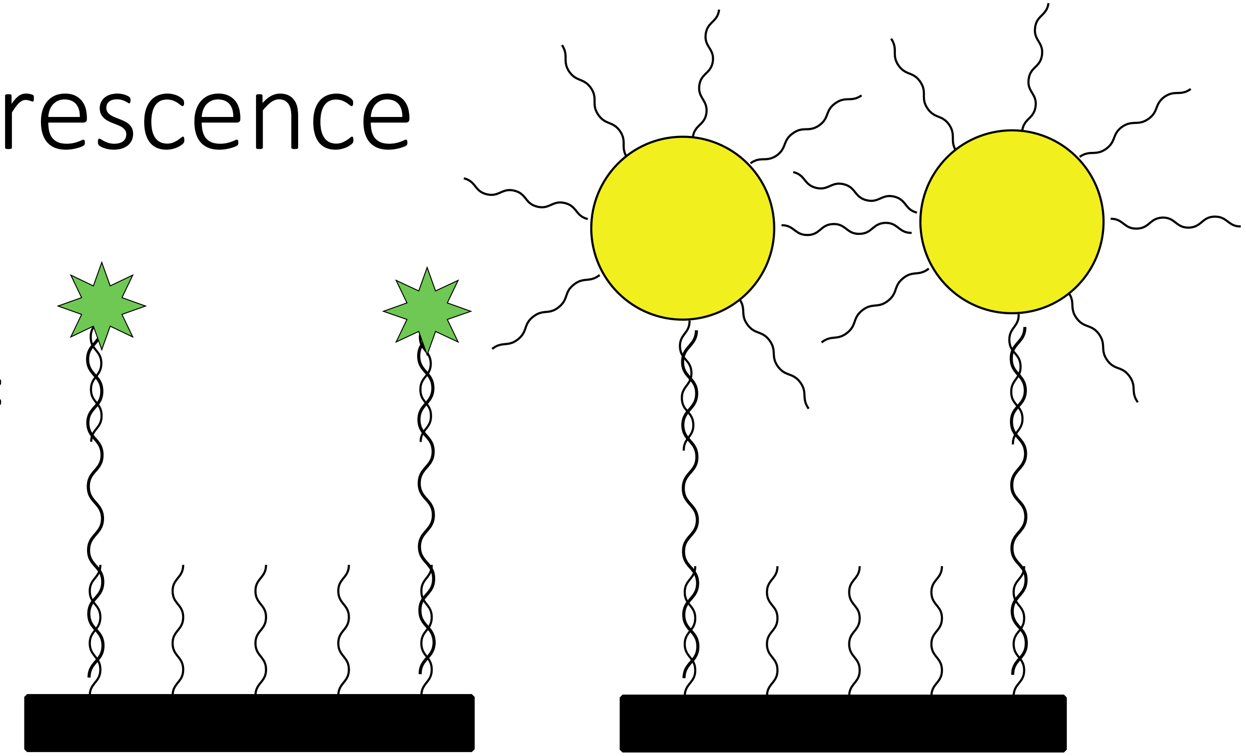
Light scattering vs. fluorescence

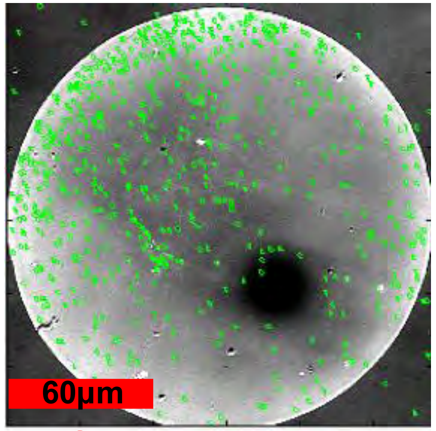
Light scattering is advantageous:

- No saturated emission rate and no photobleaching: only limit to speed is your input light power
- Inexpensive / simple instrument
- dynamic measurement capability



Replace fluorescent reporters with nanoparticle conjugates

M β-lactoglobulin



n After detection complex

LOD_{serum} < 100aM

LOD_{blood} < 1fM

analytical chemistry
Article
pubs.acs.org/ac

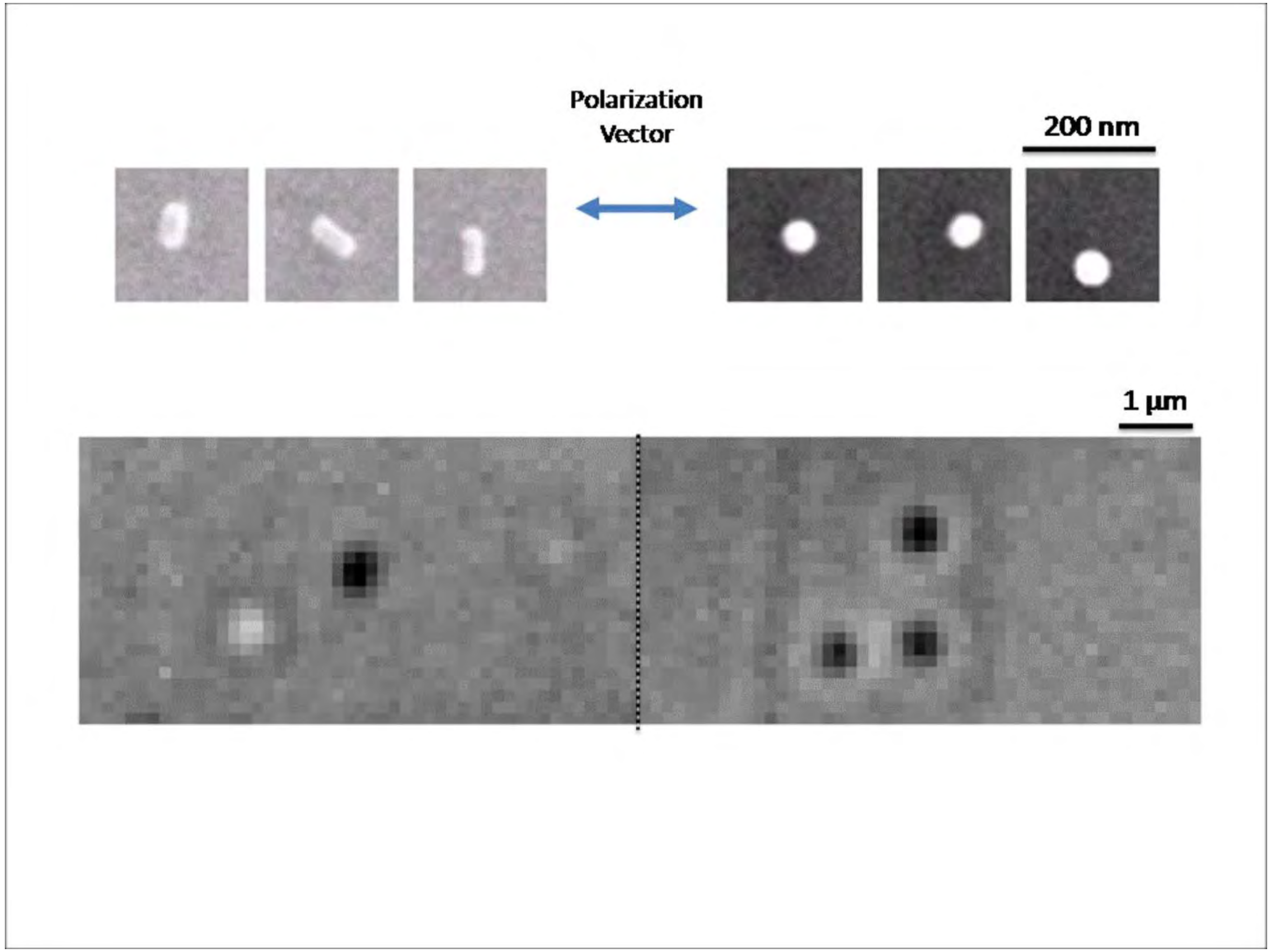
Single Nanoparticle Detection for Multiplexed Protein Diagnostics with Attomolar Sensitivity in Serum and Unprocessed Whole Blood

Margo R. Monroe,[†] George G. Daaboul,[†] Ahmet Tuysuzoglu,[‡] Carlos A. Lopez,[‡] Frédéric F. Little,[§] and M. Selim Ünlü*[,†,‡,∥]

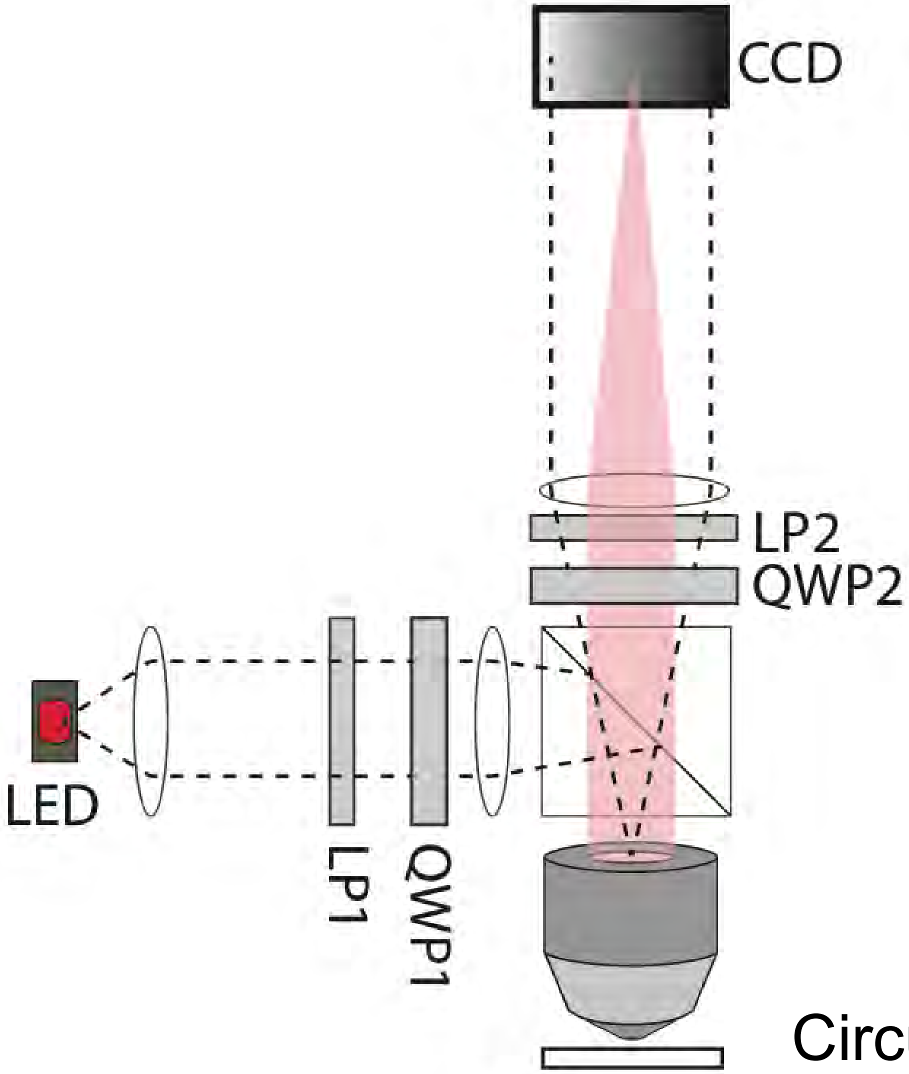
Goal: Larger field of view to scan a larger area

lower NA – lower contrast

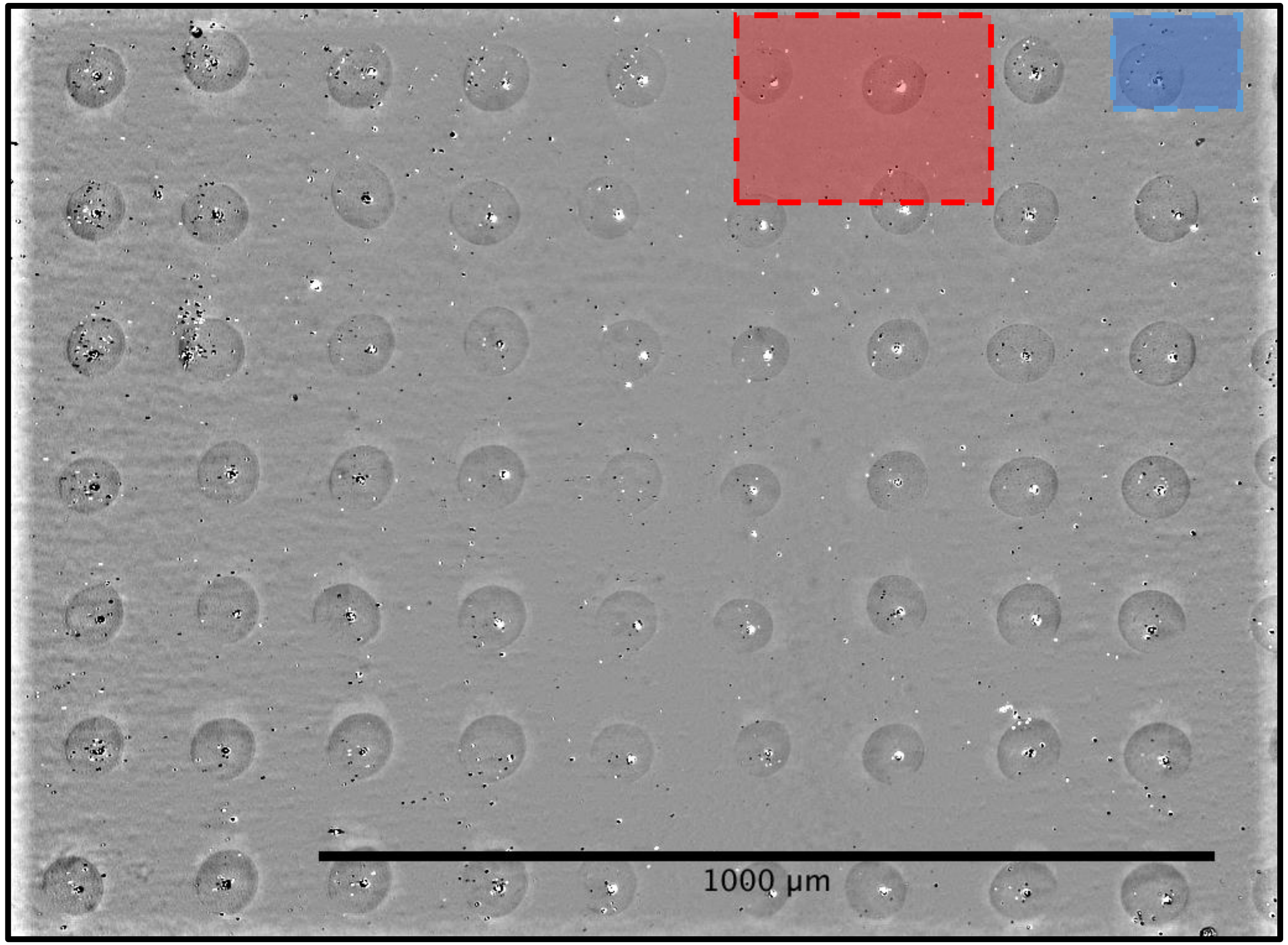
Answer: Nanorods as labels - polarization



A digital microarray with IRIS



10x Objective ... 50x ... 100x



Circular polarization enhancement

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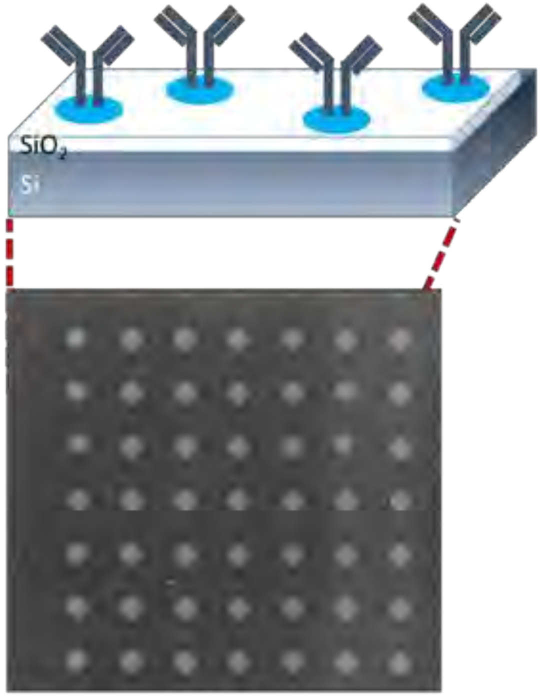
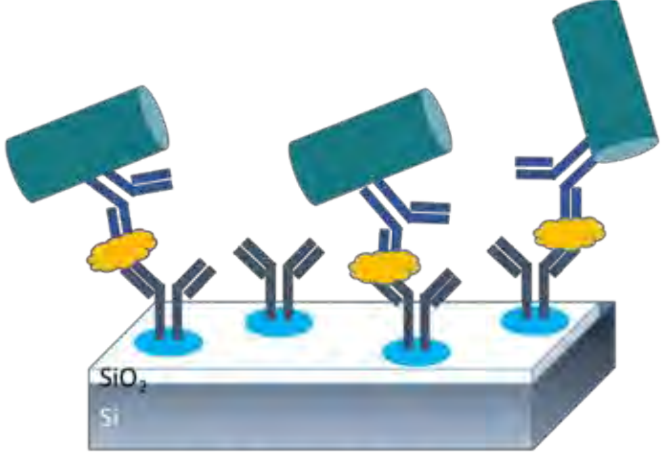
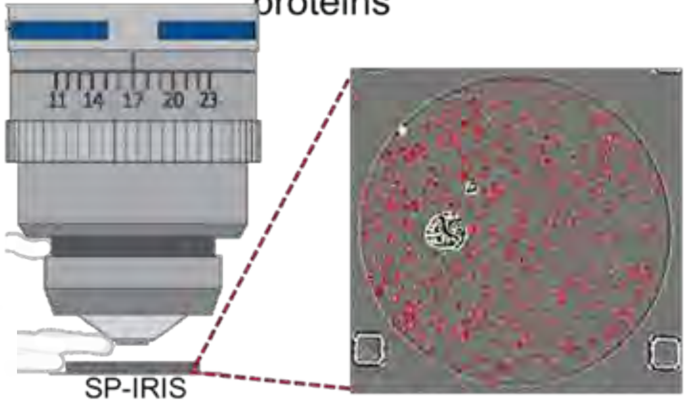
A Typical Assay



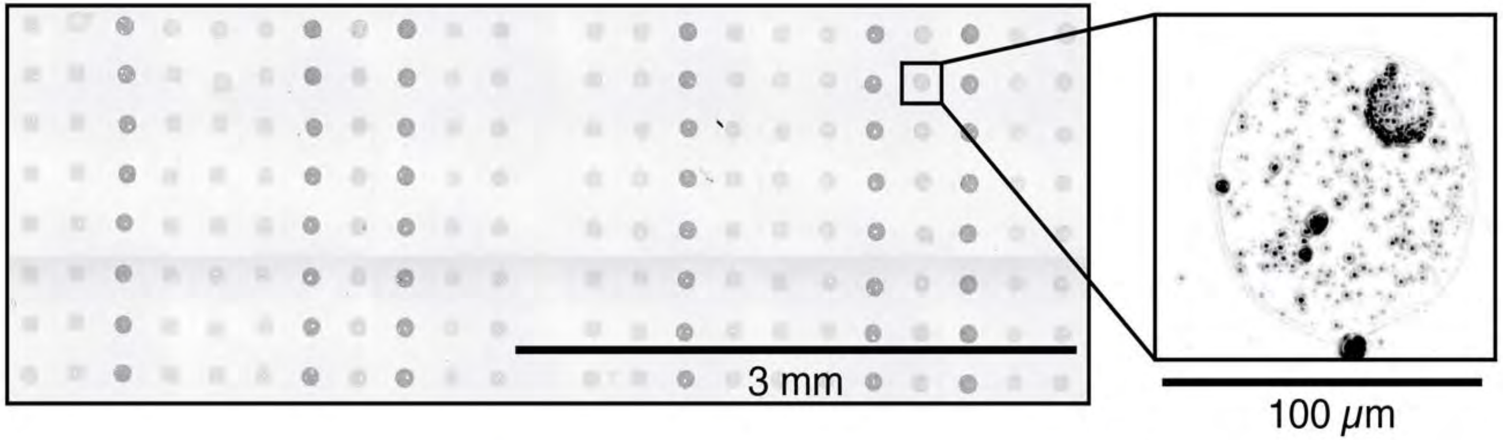
1. Antigen samples
2. Au-nanorod-tagged antibodies flow through the microfluidic array system



IRIS Single molecule detection and quantification of biomarker HBsAg proteins



Digitally detected and counted Au-nanorods captured on the spot



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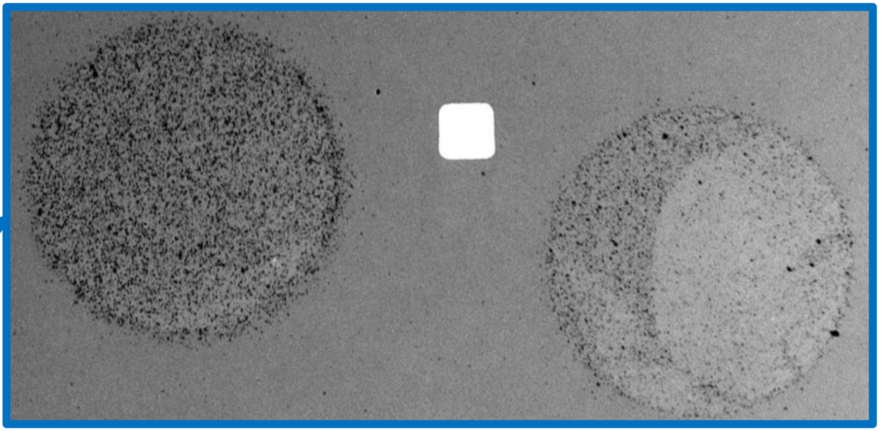
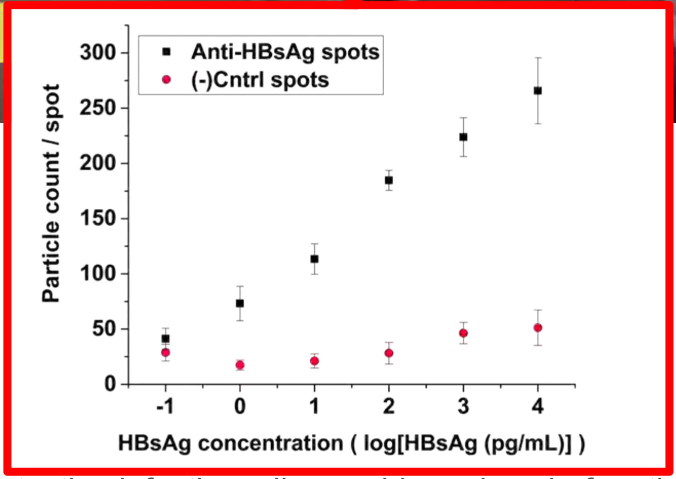
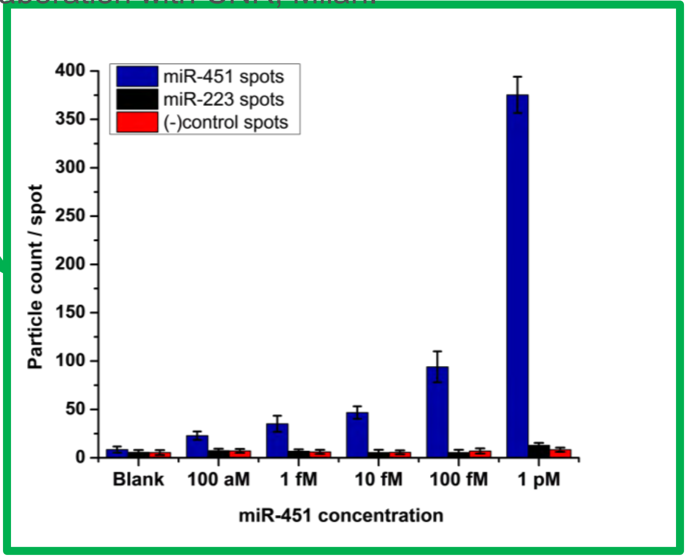
Application Areas



Detecting cancer biomarkers at ultra-low levels and rare mutations - potential to enable new options for diagnostics and treatment in cancer research. KRAS mutation detection (colorectal cancer). Work in collaboration with CNR, Milan.



Detecting infectious disease biomarkers before the onset of an immune response. HBsAg detected at 1,000X better sensitivity than commercial assays. Funded by Aselsan.



Detecting minute changes in cardiac biomarkers – at-risk patients identified earlier in their disease progression to guide more personalized care. miR-451 is a cardiac biomarker. Work in collaboration with UMass Medical.

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Commercialization

NanoView BIOSCIENCES

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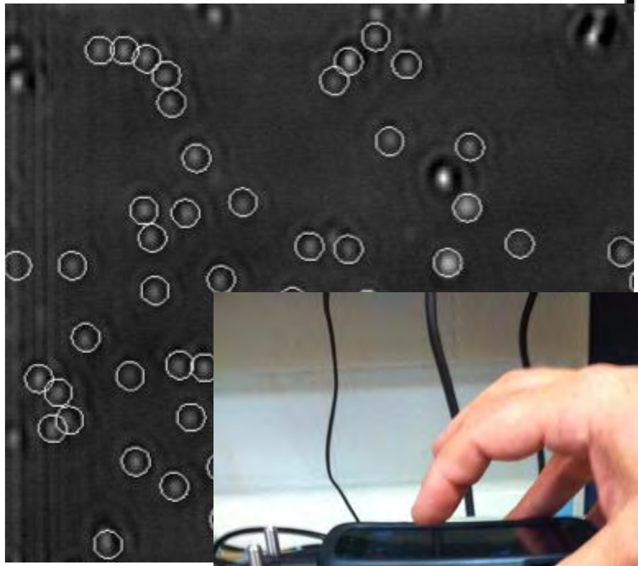
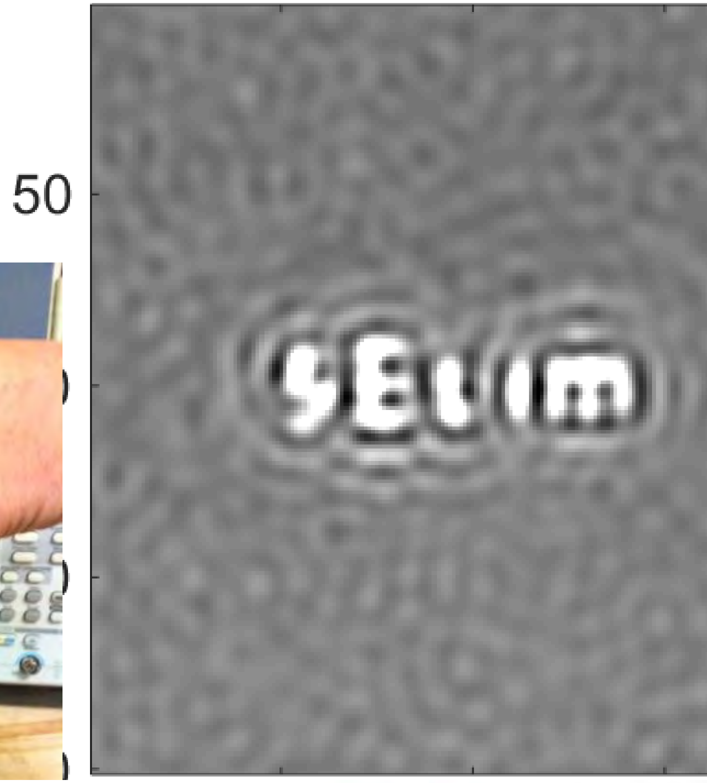
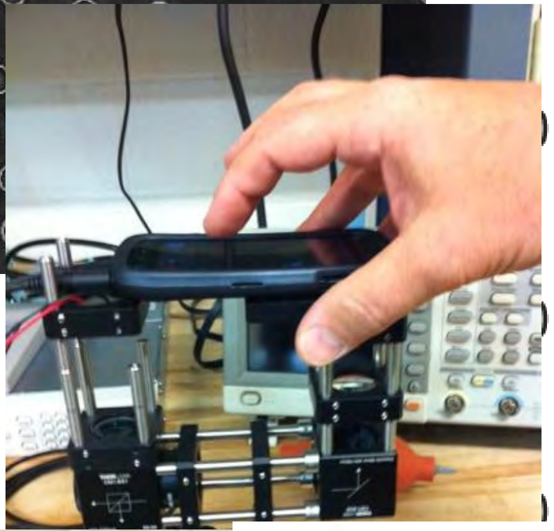
CONCLUSIONS

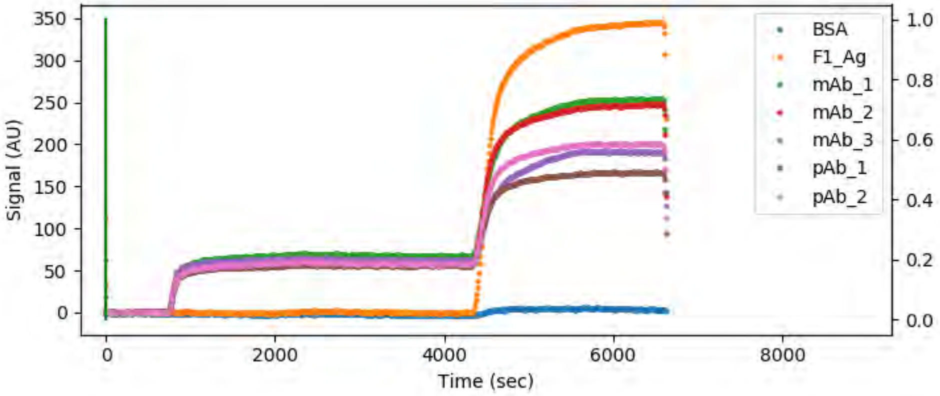
- Optical interference is a very powerful sensing technique.
- Si-chip based microfluidics for **practical** biosensing
- $10K instrument
- $5 disposables
- Multiplexed kinetic analysis
- Single biological nanoparticle detection / counting / size and shape discrimination / visualization



Reconstructed DPC image







indEx

INTEGRATED NANOPARTICLE ISOLATION AND DETECTION SYSTEM FOR COMPLETE ON-CHIP ANALYSIS OF EXOSOMES

BOSTON UNIVERSITY

NIH – NIAID, NSF
BD Biosciences
InBios, Scienion, Aselsan

Marcella Chiari (CNR, Milan)
Fred Little (MED)
John Connor (MED)

UTMB

Questions?