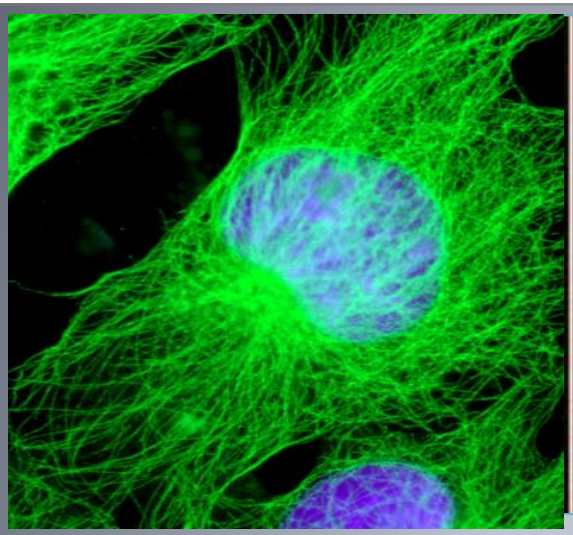
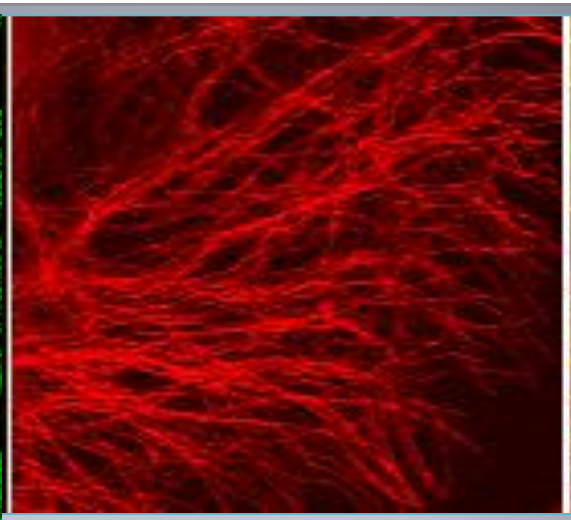
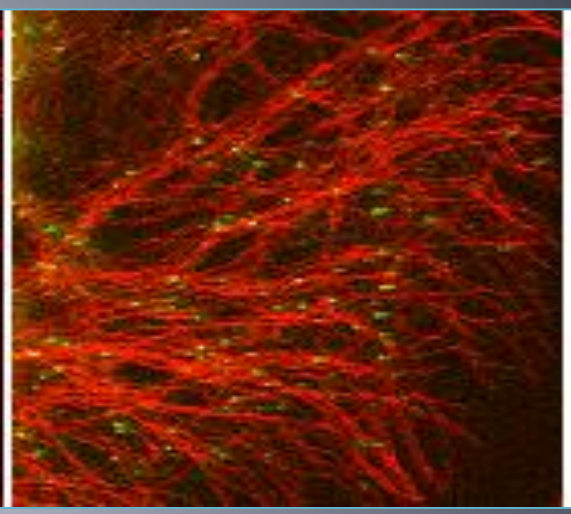
Using Circulating Tumor Cells for Precision Medicine

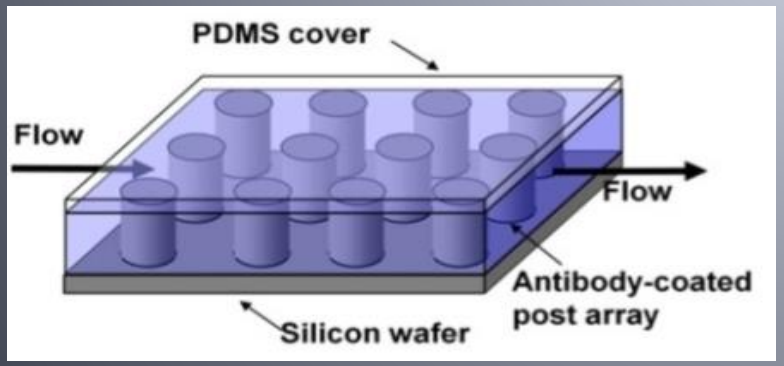




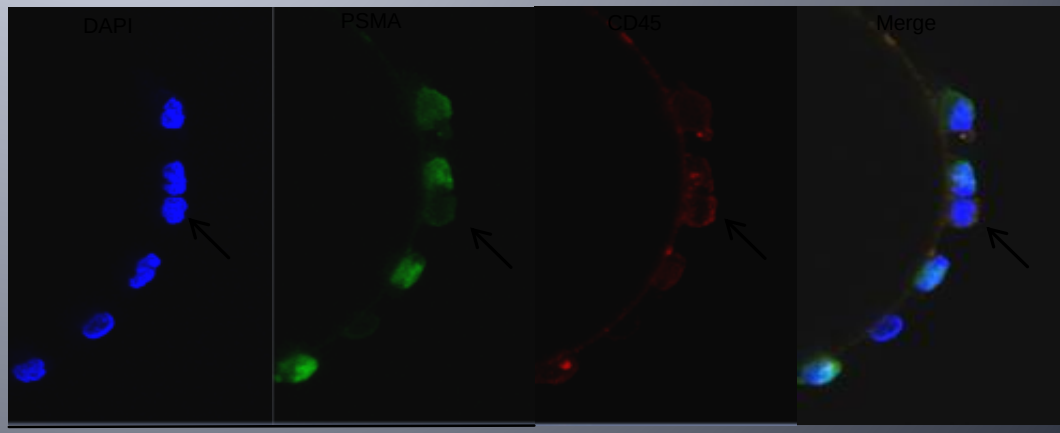


Biology

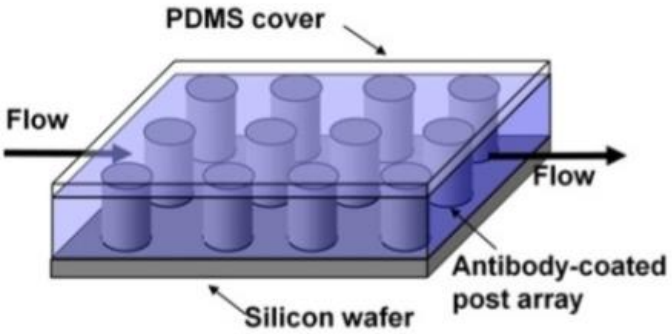
CTCs: Portal to individual patient Tumor



GEDI



The Solution: Circulating Tumor Cells

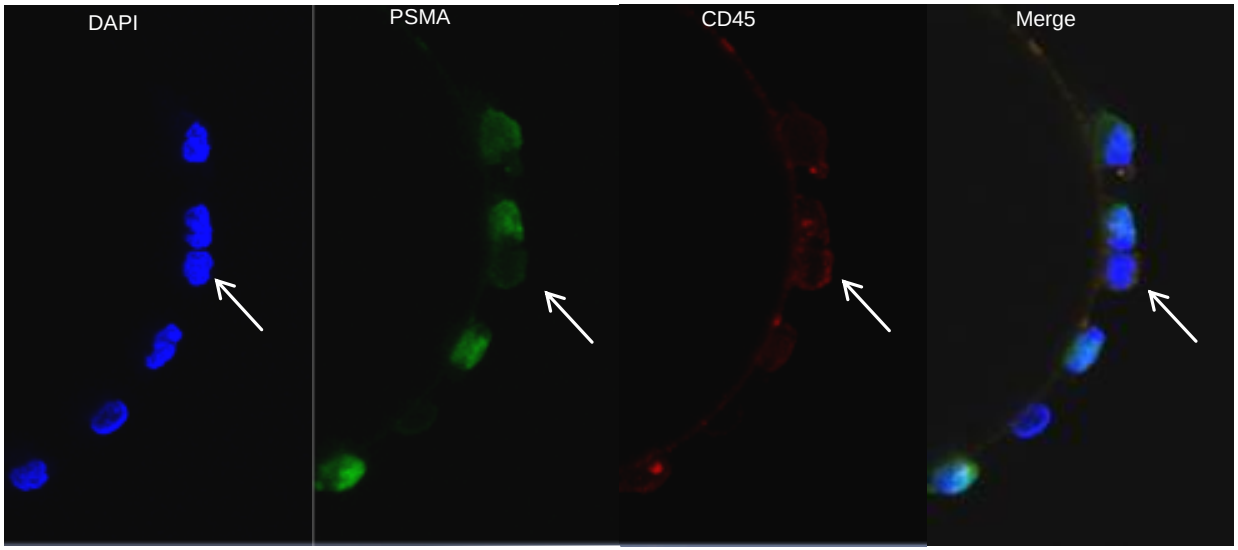


CTCs: Portal to individual patient Tumor

Brian Kirby, Engineering, Cornell, Ithaca

CTSC Pilot Funding, 2008

GEDI



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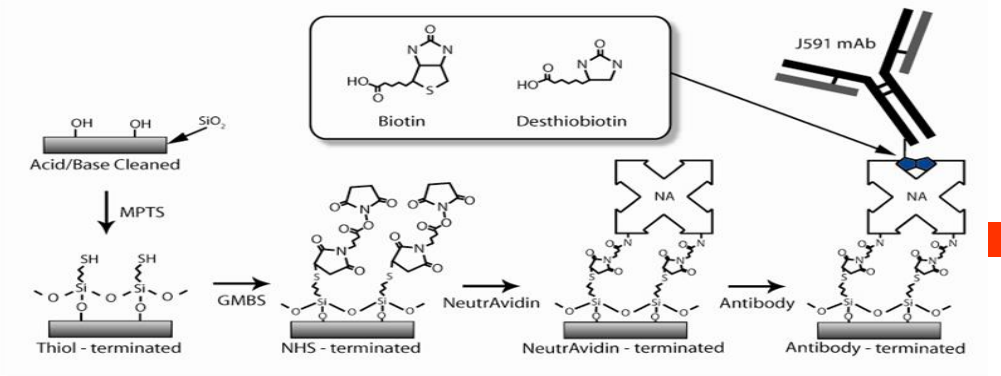
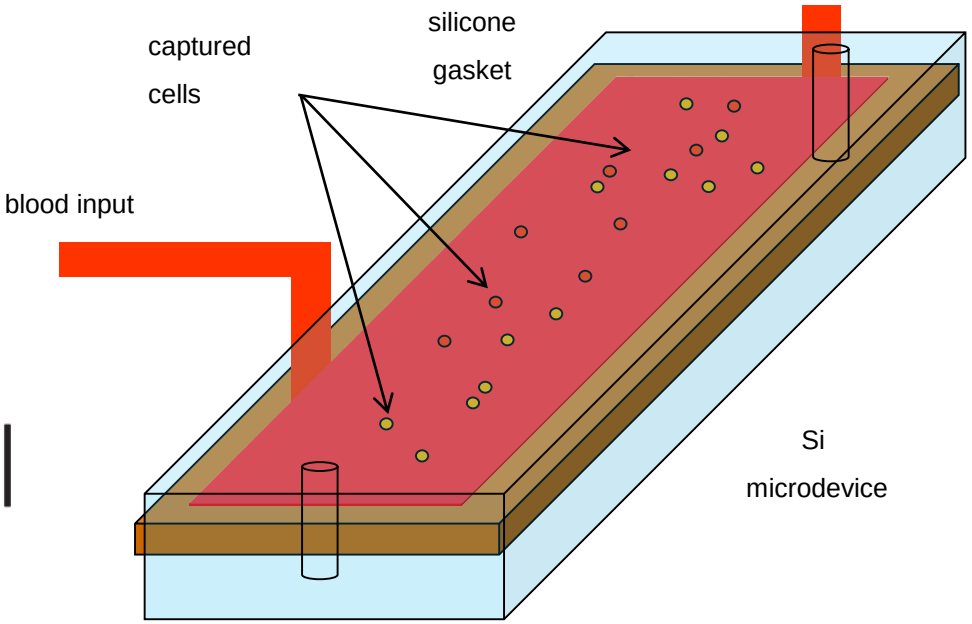
Finding a needle in a haystack: Geometrically enhanced differential immunocapture (GEDI)

- 1 mL of whole blood is injected into the device over 1 hour (flow rate at 1ml/h)
- GEDI: geometrically enhanced differential immunocapture





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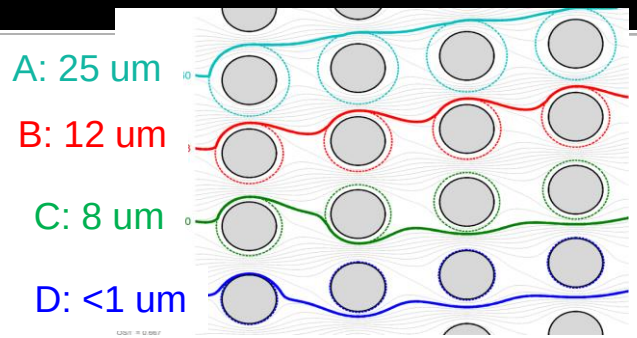
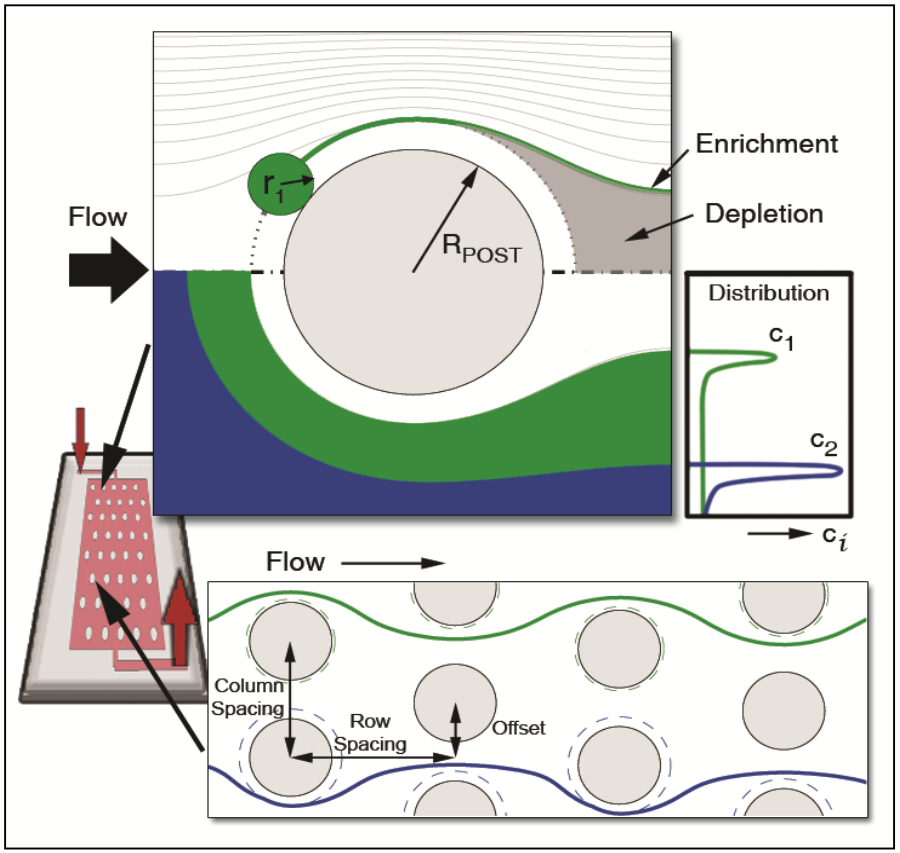
Brian Kirby and colleagues in Ithaca

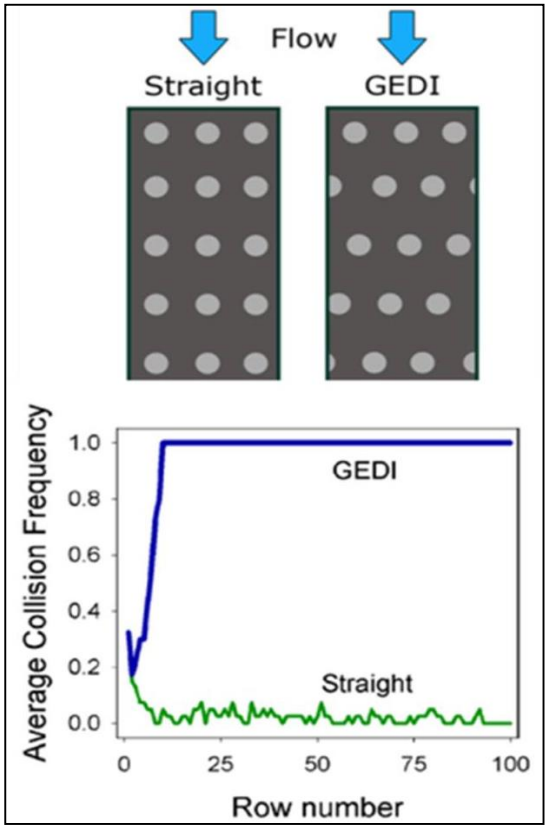
Capturing the elusive cancer cell: Fluid mechanical trickery and the development of GEDI microdevice.

- GEDI technique combines size and surface specificity to exploit differences between cancer cells and blood cells

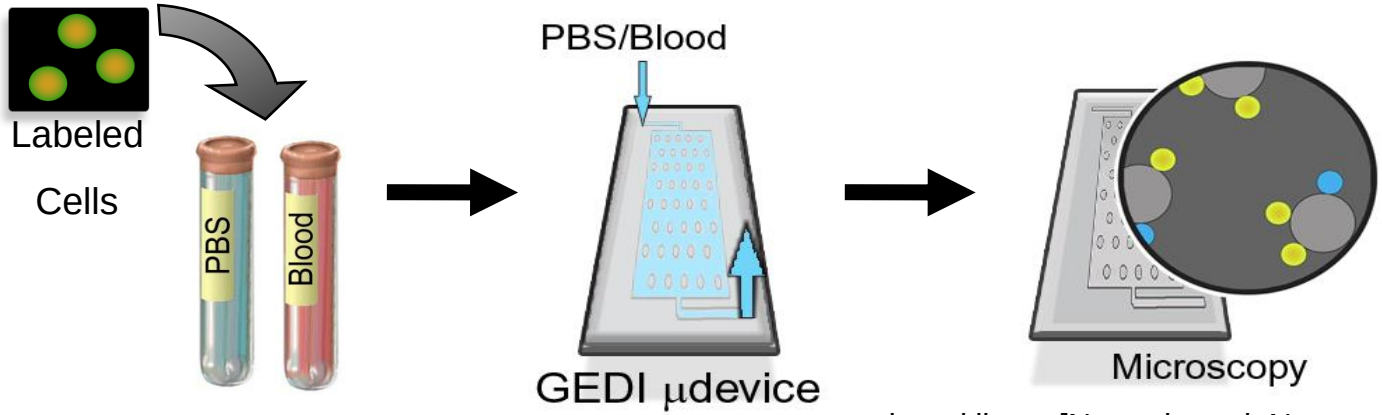


Particle trajectories through a μfluidic obstacle array.



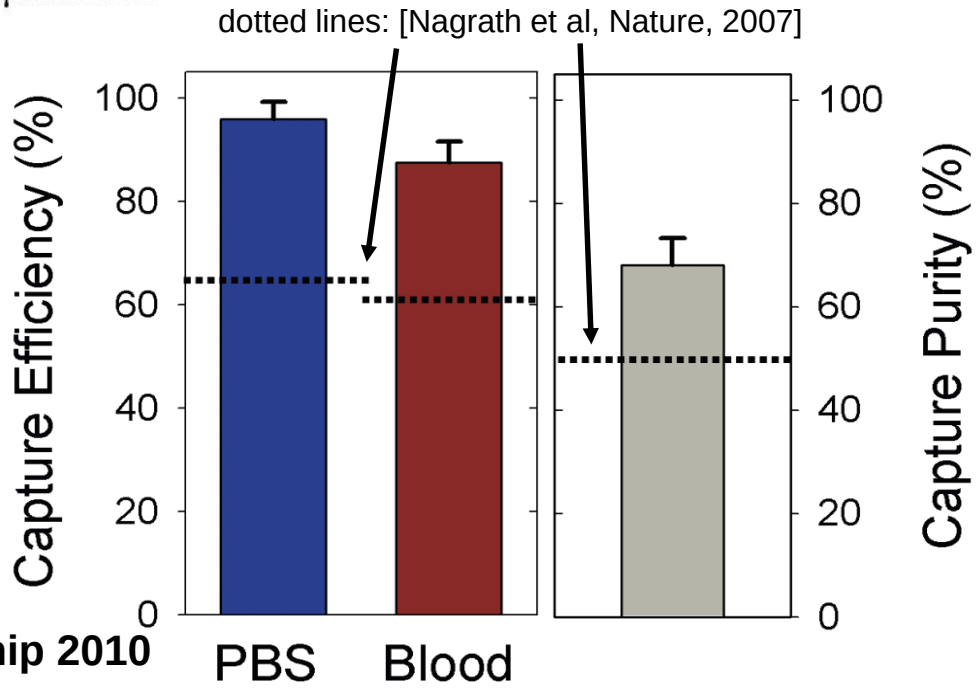


The GEDI device effectively concentrates cancer cells by a factor of 10-100 million



- 150-220 LNCaP cells/mL spiked in healthy whole blood or PBS+1% BSA

Cell type	#/mL	# captured
LNCaP	200	170
Blood	5 billion	91



Gleghorn, Giannakakou, Kirby et al **Lab on a Chip 2010**

Timeline of GEDI Funding

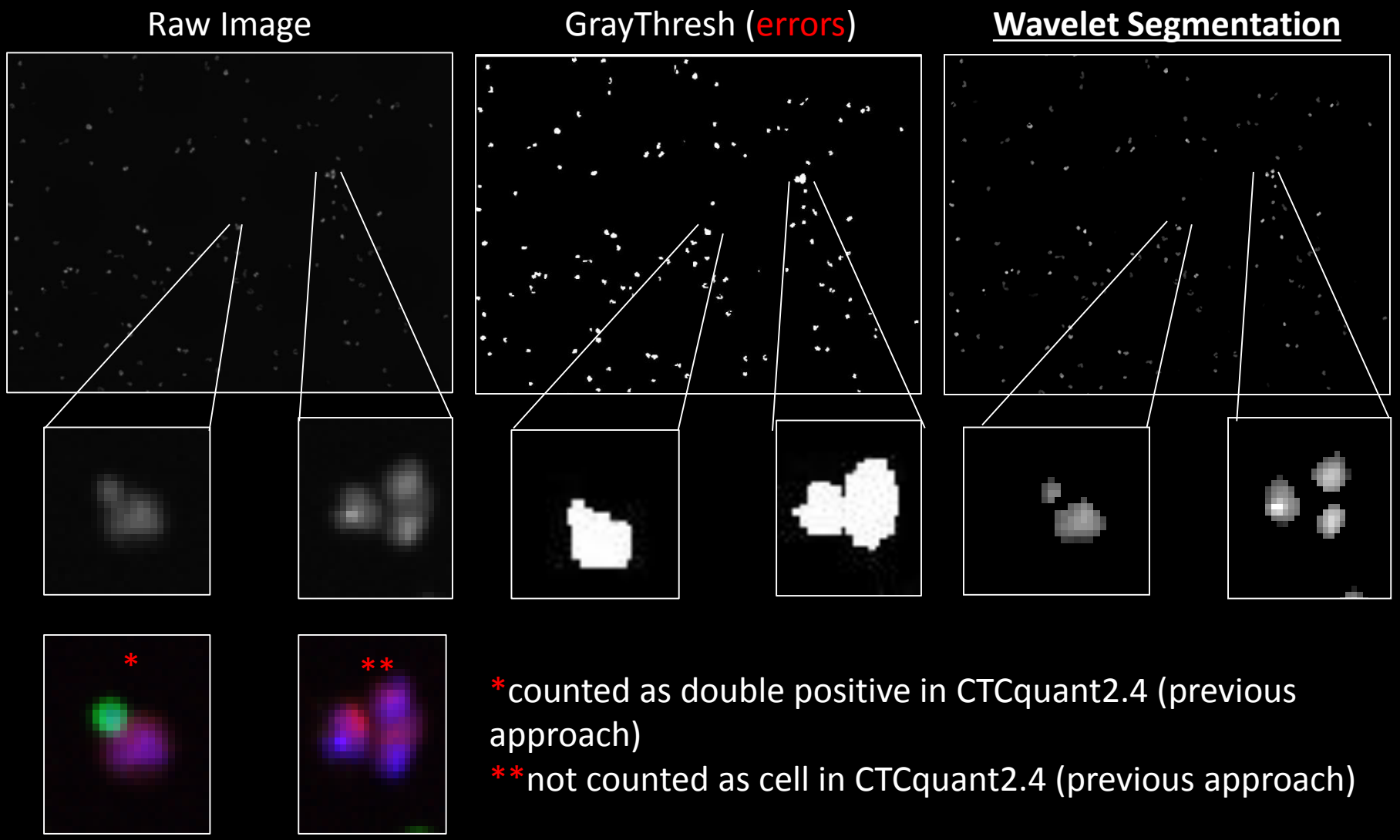
- Partnership with Brian Kirby, Engineering, Cornell, Ithaca
- **2008, CTSC Pilot Funding**
- 2009, NIH R01 (Giannakakou; Nanus)
- 2009, NYSTAR Designated Center for Advanced Technology (Kirby; Giannakakou)
- 2009, PCF Creativity Award (Nanus; Giannakakou; Kirby)
- 2011, NIH Center on the Microenvironment and Metastasis (Giannakakou;Project 2 co-leader; Kirby GEDI nanofabrifaction, Nanus, Project 4 co-leader)
- Sanofi, Prospective Phase II Clinical Trial in CRPC, 2013

- 2012, Development of HER2-GEDI for Breast and Gastric Cancer
- 2012, MWCBC, Breast Cancer Grant (Giannakakou)
- 2012, Eisai Pharma Breast cancer study (Giannakakou)

- 2013, Alex Matov, NRSA postdoctoral fellowship
- **2013, Alex Matov, CTSC Pilot grant**

Wavelet-based DAPI Segmentation (Incubation chip pos350)

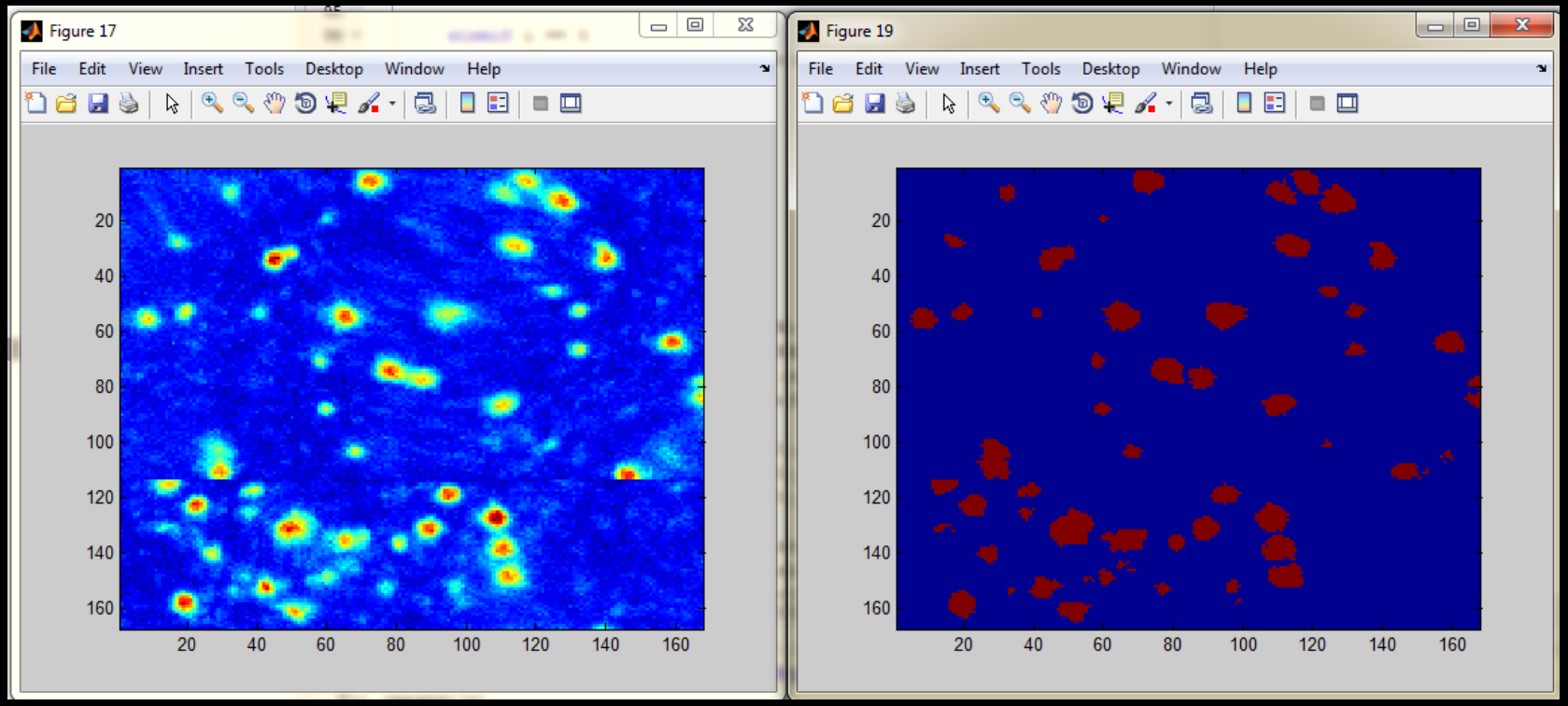
cells merged with earlier algorithm, which was erroneous



*counted as double positive in CTCquant2.4 (previous approach)

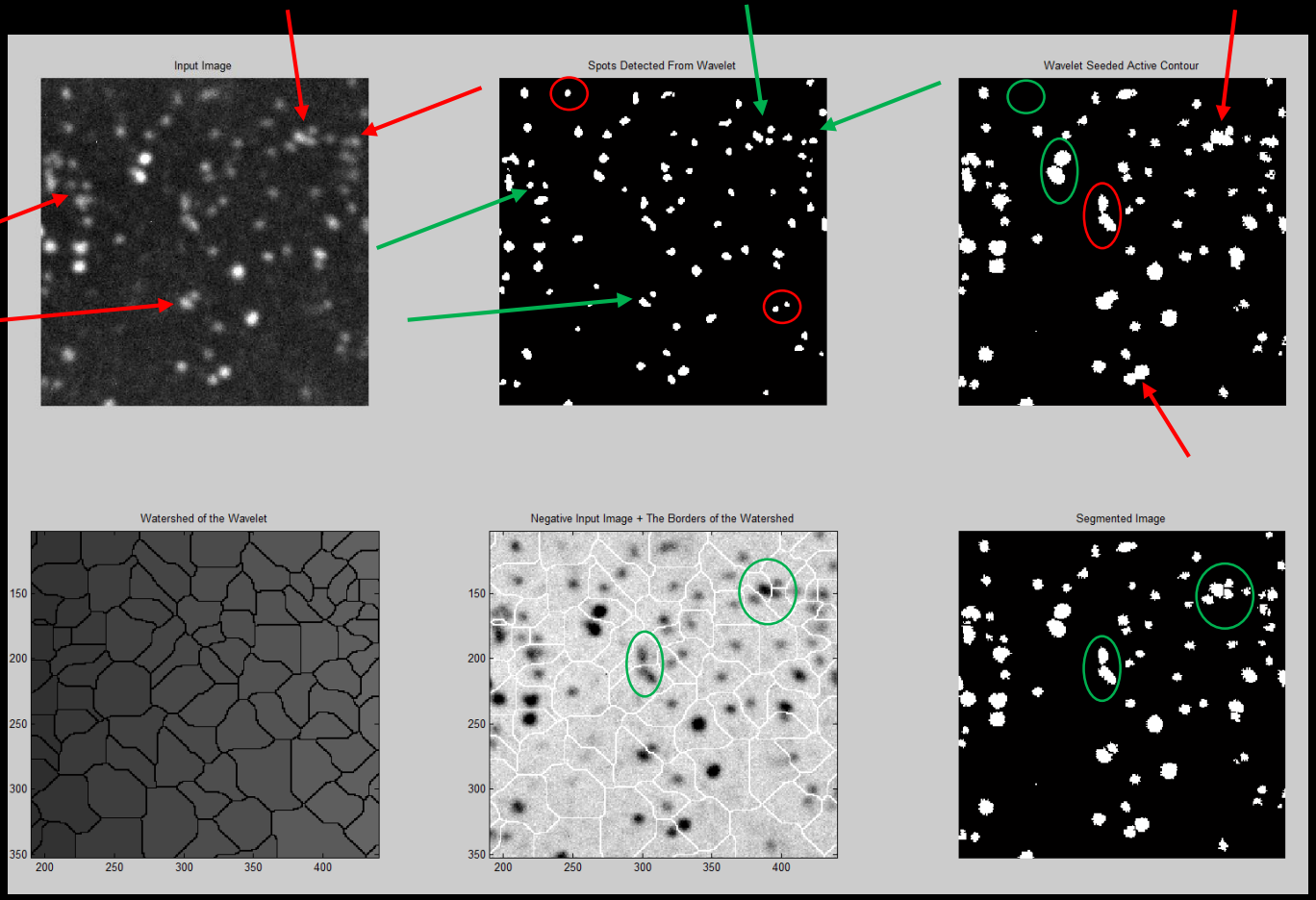
**not counted as cell in CTCquant2.4 (previous approach)

New segmentation method based on wavelet seeding and active contour using watershed limits of the area



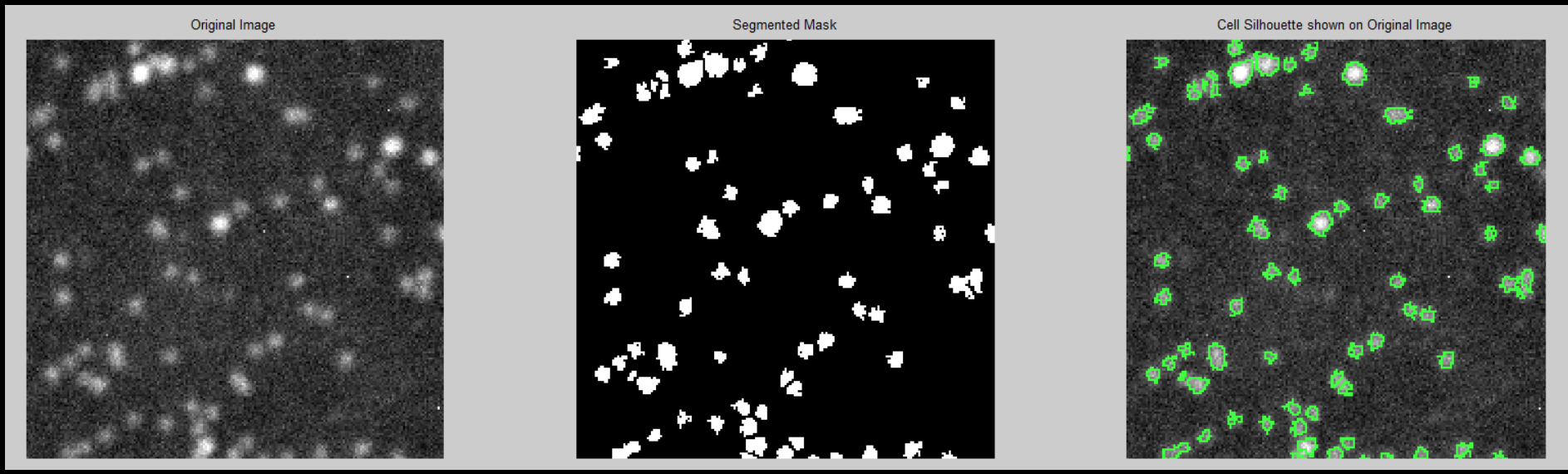
Algorithm designed and initiated by Alex Matov, and implemented in Matlab by Shayan Modiri (a student hired by A. Matov to work on his NCATS-NIH grant)

Novel PSMA Segmentation Method (Matov)



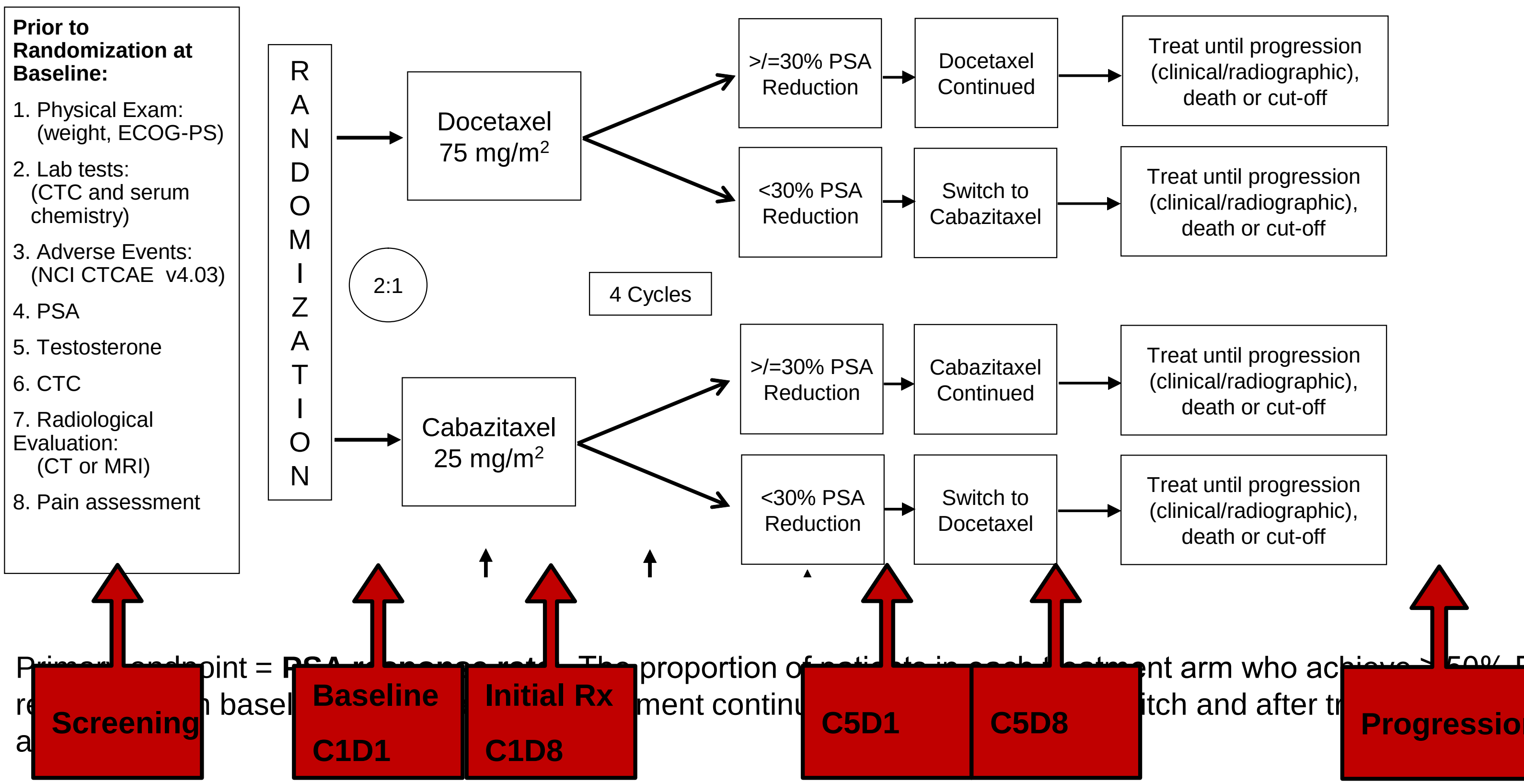
The areas of adjacent cells are correctly segmented even if the cells partially overlap

Novel PSMA Segmentation Method (Matov)



- The initial image segmentation is accomplished by stationary wavelet transform, which identifies bright pixel clusters in noisy images (seeding step)
- Active contour, next, identifies precisely the edges of the image features based on the seeds
- Watershed transformation of the seeding step image is overlaid, with reversed intensities, on the active contour image; logical conjunction ('and') of the active contour image and the watershed image identifies the bright areas and their exact borders

TAXYNERGY: Phase II Trial to Evaluate Benefit of Early Switch from first-Line Docetaxel/Prednisone to Cabazitaxel/Prednisone and the opposite sequence, exploring molecular markers and mechanisms of taxane resistance in men with Metastatic Castration-Resistant Prostate Cancer (mCRPC) who have not received prior chemotherapy



Prior to Randomization at Baseline:
1. Physical Exam: (weight, ECOG-PS)
2. Lab tests: (CTC and serum chemistry)
3. Adverse Events: (NCI CTCAE v4.03)
4. PSA
5. Testosterone
6. CTC
7. Radiological Evaluation: (CT or MRI)
8. Pain assessment
RANDOMIZATION
2:1
Docetaxel 75 mg/m^2
Cabazitaxel 25 mg/m^2
4 Cycles
>/=30% PSA Reduction
Docetaxel Continued
Treat until progression (clinical/radiographic), death or cut-off
<30% PSA Reduction
Switch to Cabazitaxel
Treat until progression (clinical/radiographic), death or cut-off
>/=30% PSA Reduction
Cabazitaxel Continued
Treat until progression (clinical/radiographic), death or cut-off
<30% PSA Reduction
Switch to Docetaxel
Treat until progression (clinical/radiographic), death or cut-off
Screening
Baseline C1D1
Initial Rx C1D8
C5D1
C5D8
Progression

Pr… endpoint = **PSA r…** The proportion of … in each t…ment arm who ac… …50% PSA r… basel… …ment continu… …itch and after tr…n, if a…

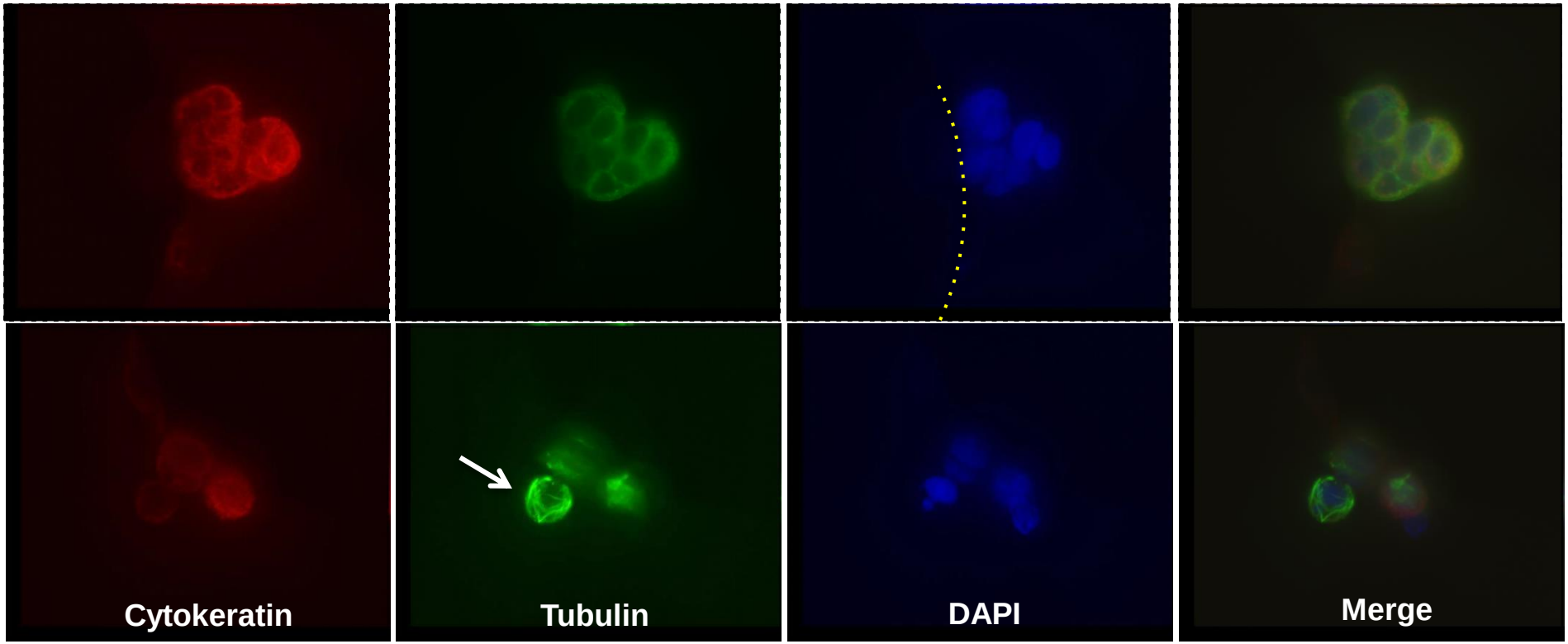
Co-Primary endpoint = **Drug Target Engagement**: The potential prognostic value of prostate cancer circulating tumor cells will be determined by using CTCs isolated at baseline and at defined time-points using the geometrically enhanced differential immunocapture (GEDI) method; by performing CTC enumeration, AR localization (cytoplasmic and nuclear) and tubulin bundling. The findings will be correlated with the clinical response/resistance to docetaxel/cabazitaxel for all patients

Clinical co-chairs: Antonarkis, Tagawa

Biomarker chair: Giannakakou

Assays on the Chip
4. Ex Vivo Treatment of LIVE CTCs to Predict Chemotherapy Response

Captured CTCs Ex Vivo treatment with Different Taxanes: Can CTC Response Predict Clinical Response?



Dotted Line: micropost margins

Arrow head: intricate microtubule network

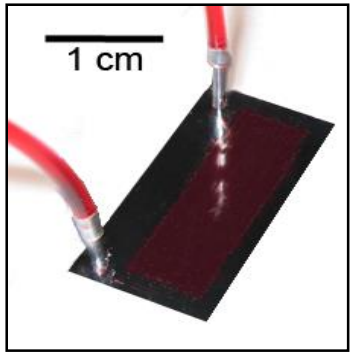
Arrow: bundled microtubule (DTE effect)

Customize Chemotherapy for the Individual

We aim to facilitate personalized treatment via capture and analysis of rare cells

Patient Sample

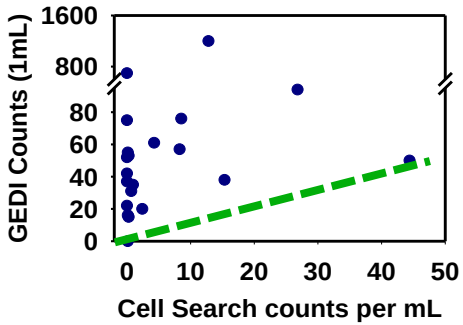
Cancer cell Capture



- our unique contribution: viable CTCs on chip for FUNCTIONAL ANALYSIS

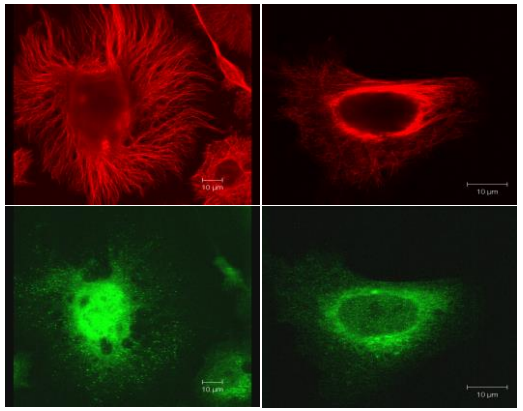
Enumeration/

Genomics



Functional

Response



- Can we predict the response of prostate cancer patients to chemotherapy?
- Can we Tailor Therapy in Real Time?

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- Nancy Chan, MD
- Sidharta Shen, PhD
- Marisa Carbonaro, PhD
- Alex Matov, PhD

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- **Scott Tagawa, MD (Weill Cornell)**
- **Neil Bander, MD (Weill Cornell)**
- **Brian Kirby, PhD (Cornell University)**
- **Stephen Plymate, MD (University of Washington, Seattle)**

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  - Jason Gleghorn, Phd
  - Erica Pratt, PhD candidate
    - Yusef Syed
    - Erica Pratt
    - Steven Santana
    - Tim Lannin
    - Jim Smith
    - Charlie Huang