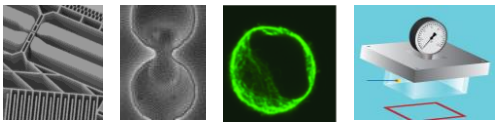
Cell mechanotype:
from screening to disease biophysics



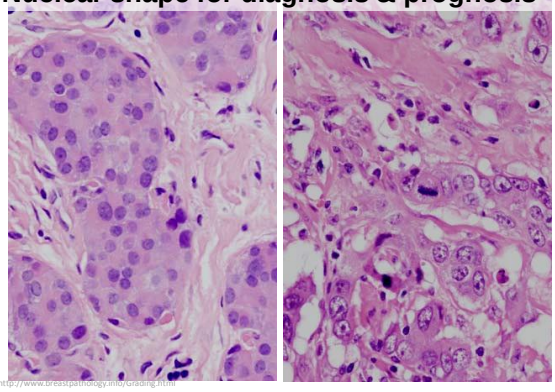
Amy Rowat
Dept of Integrative Biology and Physiology, UCLA

Tumor stiffness for detection

- Physical palpation to detect malignancy

wiley-vch.e-bookshelf.de

Nuclear shape for diagnosis & prognosis



http://www.breastpathology.info/grading.html

Cancer cell physical properties

Nuclear Shape | Adhesion

Cytoskeletal Organization | Traction Stresses

Deformability

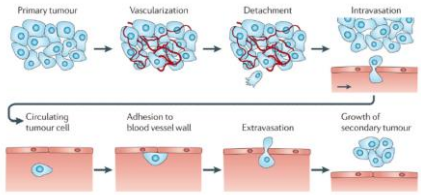
Wirtz et al (2012) *Nat Rev Cancer*

Cancer cell physical properties

Nuclear Shape | Adhesion

Cytoskeletal Organization | Traction Stresses

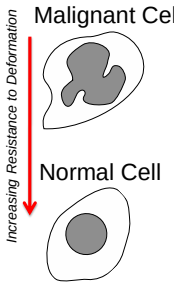
Deformability

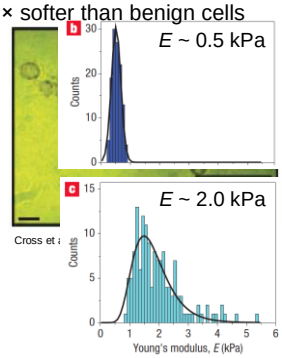


Wirtz et al (2012) *Nat Rev Cancer*

Cancer cells are softer

- Malignant cells ~ 2 to 5 × softer than benign cells



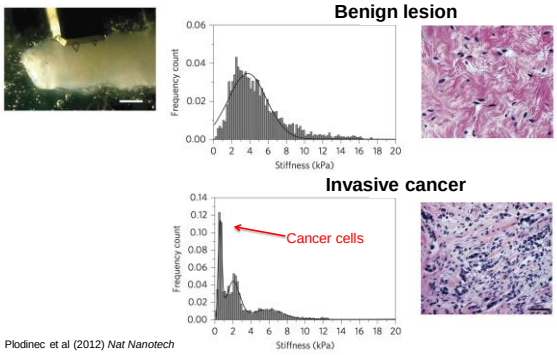


Cross et a

Xu et al (2012) *PLoS ONE*; Swaminathan et al (2011) *Cancer Res*

Cancer cells are softer

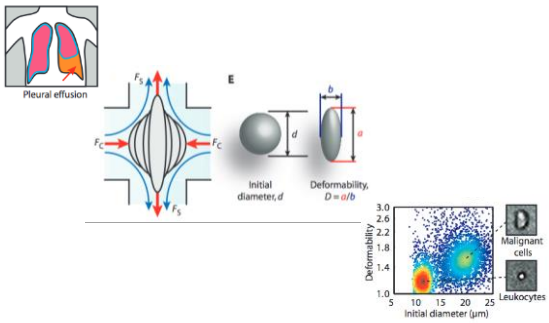
- Human breast biopsies have distinct stiffness profiles



Plodinec et al (2012) *Nat Nanotech*

Mechanotype as biomarker

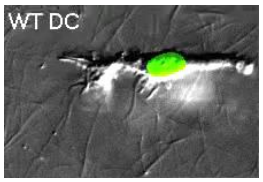
- Improve clinical decision-making



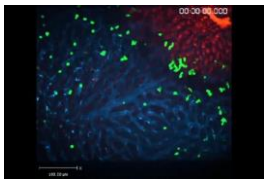
Tse et al (2013) *Sci Transl Med*

Cell mechanical properties matter

Cells need to deform



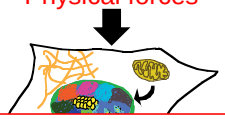
Lämmermann et al (2008) *Nature*



McDonald et al (2010) *Science*

Cell mechanical properties matter

Physical forces



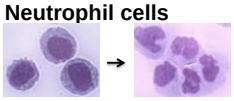
→ Understanding the interplay between physical forces and biochemical processes is a major challenge in modern cell biology

Rowat Lab

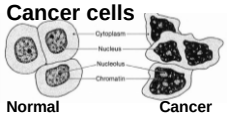
Mechanics <-> Physiology <-> Disease

The physical & molecular origins of cell/ nucleus shape and stability

Neutrophil cells



Cancer cells



Normal **Cancer**

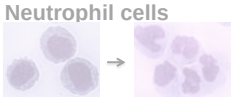
Rowat Lab

Mechanics <-> Physiology <-> Disease

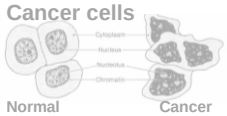
The physical & molecular origins of cell/ nucleus shape and stability

Technologies for probing cell/ nuclear mechanical properties with higher throughput

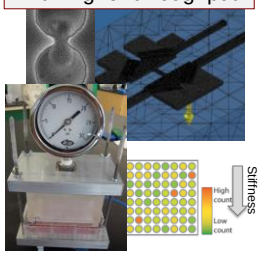
Neutrophil cells



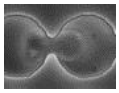
Cancer cells



Normal Cancer



Mechanotype in cancer

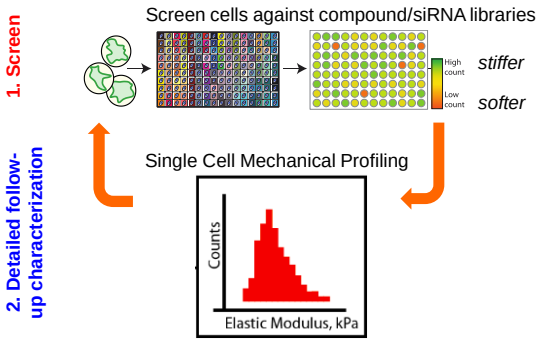


The Mechanotyping Platform

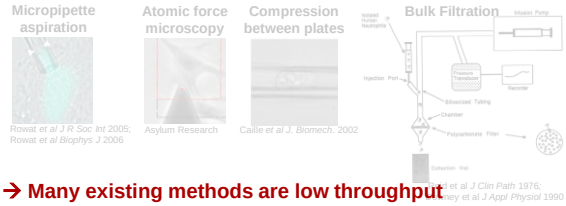
Molecular origins of mechanotype

Mechanotype in cancer progression

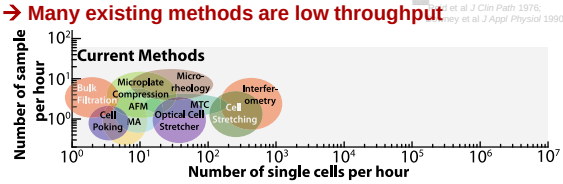
Screen based on mechanotype



Measuring mechanotype of cells and nuclei

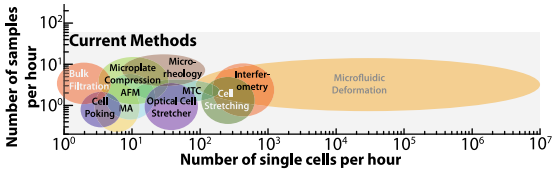


→ **Many existing methods are low throughput**

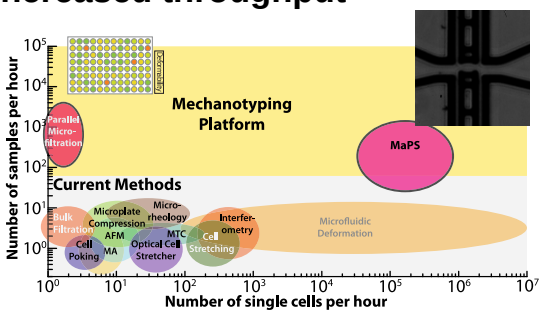


Measuring mechanotype of cells and nuclei

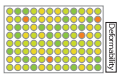
→ **Microfluidic methods rely on imaging; challenging to scale up**

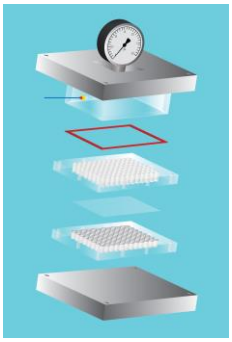


Goal: Mechanotyping with increased throughput



Parallel microfiltration





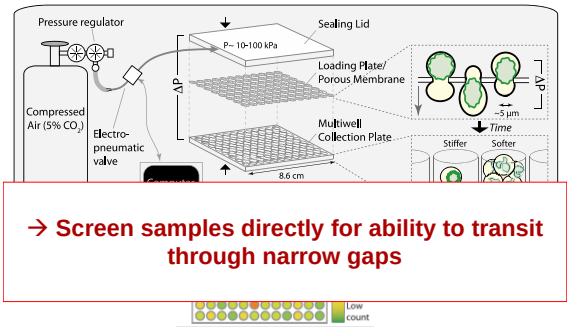
Dongping Qi, PhD,
Postdoctoral Researcher

Navjot Kaur Gill
Graduate Student Researcher

Rowat & Qi, Patent WO/2013/056253; Qi*, Gill*…Rowat, Nat Sci Reports, *Revisions requested*

Parallel microfiltration

• Perform hundreds of deformation assays in parallel

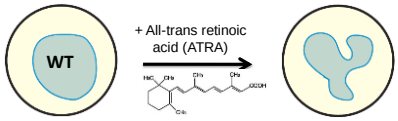


→ **Screen samples directly for ability to transit through narrow gaps**

Model system for validation

HL-60
(Wildtype)

ATRA-treated HL-60
(Neutrophil-type)



→ Cells have different mechanotype:

E ~ 900 Pa

E ~ 150 Pa

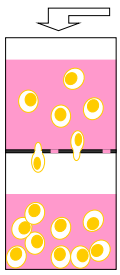
→ Cells have similar size distributions

*Rosenbluth et al *Biophys J* 2006

Parallel microfiltration

Pressure

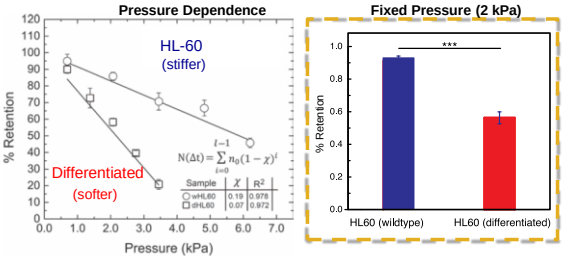
Softer



Stiffer

Mechanotyping by microfiltration

• Proof of principle – distinguish between cell types:
HL-60 (Wildtype) vs. Neutrophil-type (Differentiated)

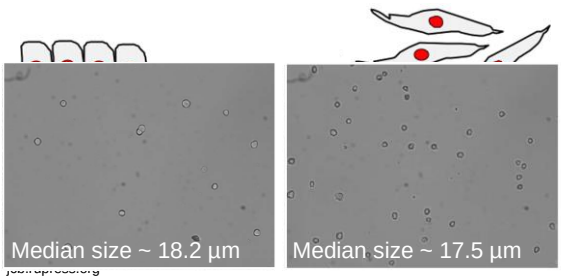


8 µm pores; 50 sec; N>15 wells over 3 independent experiments
Qi*, Gill*…Rowat, Nat Sci Reports, *Revisions requested*

Mechanotyping by microfiltration

Epithelial
Less motile
Stronger cell-cell interactions
+ E-Cadherin

Mesenchymal
Motile
Invasive
- E-Cadherin



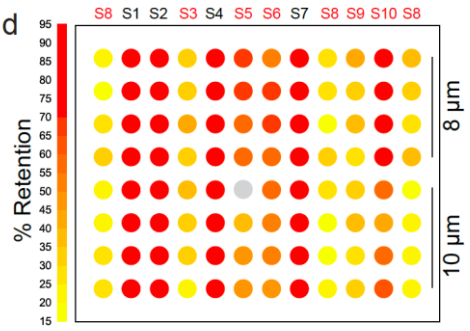
PMF-screen panel of ovarian cancer cells

• Panel of ovarian cancer cells derived from mouse and human
• Transformed to mesenchymal-type through distinct mechanisms

With Ruprecht Wiedemeyer, Cedars Sinai

Qi*, Gill*…Rowat, Nat Sci Reports, *Revisions requested*

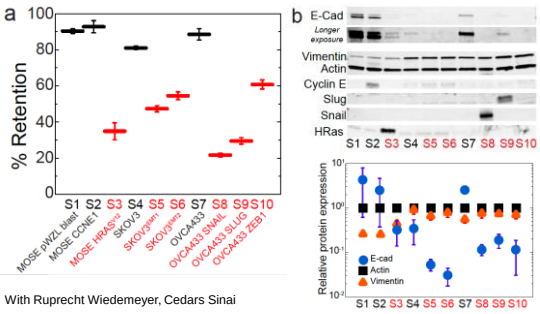
PMF-screen panel of ovarian cancer cells



Qi*, Gill*…Rowat, Nat Sci Reports, *Revisions requested*

Mesenchymal-type cells are softer

- Panel of ovarian cancer cells derived from mouse and human
- Transformed to mesenchymal-type through distinct mechanisms



With Ruprecht Wiedemeyer, Cedars Sinai

Qi*, Gill*…Rowat, Nat Sci Reports, *Revisions requested*

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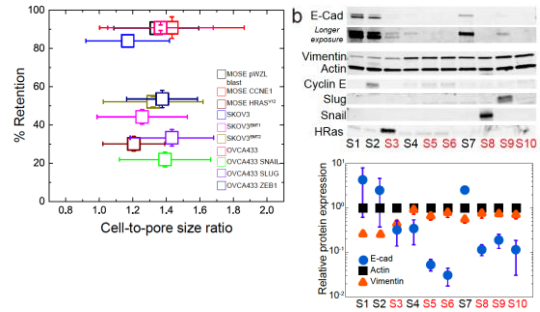
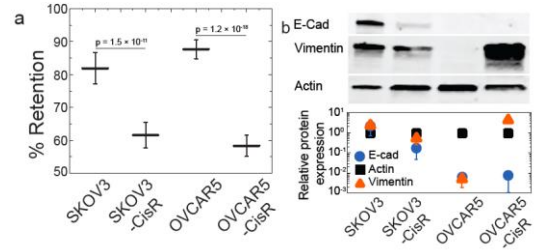


Qi*, Gill*…Rowat, Nat Sci Reports, *Revisions requested*

Drug resistant cells are softer

• Cisplatin-resistant cells are softer; have mesenchymal phenotype/mechanotype

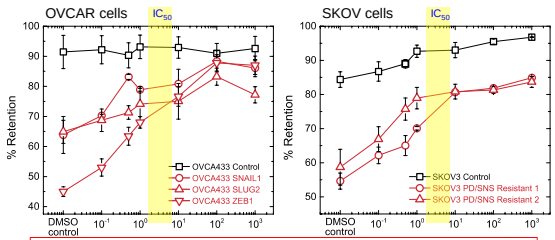


With Jianyu Rao, UCLA & Oliver Dorigo, Stanford

Qi*, Gill*…Rowat, Nat Sci Reports, *Revisions requested*

Drug response: mesenchymal vs. epithelial cells

• Treatment with chemotherapy drug, paclitaxel

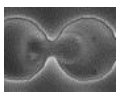


→ **Screen for anti-cancer compounds**

Qi*, Gill*…Rowat, Nat Sci Reports, *Revisions requested*

Liebmann et al. (1993) *Brit J Cancer*

Mechanotype in cancer



The Mechanotyping Platform:
→ Screen based on mechanotype.
Detect EMT based on mechanotype.

What are the molecules that regulate mechanotype?

Mechanotype in cancer progression

Acknowledgements

CAREER Award

UCLA Physical Sciences Entrepreneurship & Innovation Award

Mesenchymal-type cells are softer

- Single cells occlude pores; no clustering