Towards Patient Specific Organotypic Models of Cancer

mmb LAB

DEPARTMENT OF Biomedical Engineering
College of Engineering University of Wisconsin-Madison

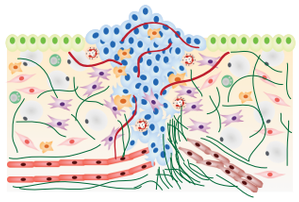
Carbone Cancer Center
UNIVERSITY OF WISCONSIN
SCHOOL OF MEDICINE AND PUBLIC HEALTH

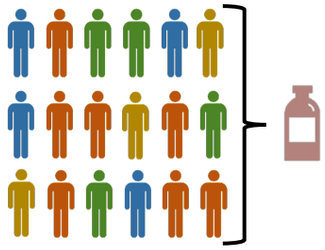
DEPARTMENT OF PATHOLOGY AND LABORATORY MEDICINE
University of Wisconsin School of Medicine and Public Health

Disclaimer: D.J. Beebe has equity in Bellbrook Labs LLC, Tasso, Inc., Stacks to the Future LLC, Lynx Biosciences, LLC, Onexio Biosystems LLC, Turba LLC, and Salus Discovery LLC.

Key Points

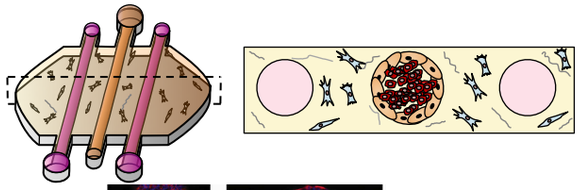
- **Cancer & Clinical Decision Making**

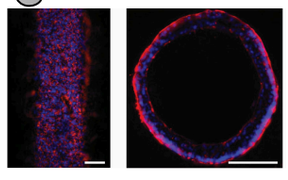


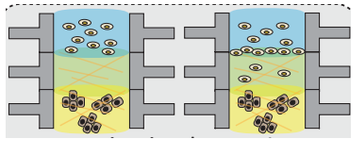


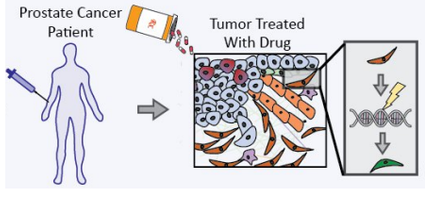
- **"Organotypic" Models of Cancer**

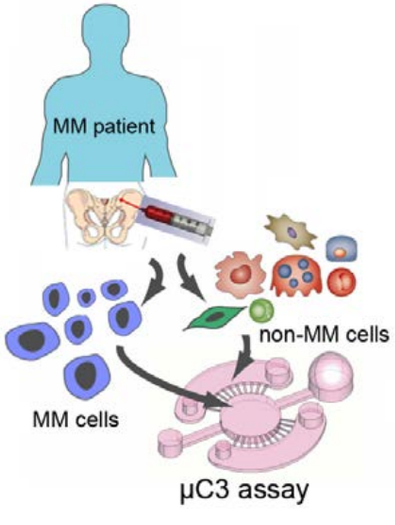
  - ➡ ***"Better" biology via structure & microenvironment***
  - ➡ ***How much is enough?***
  - ➡ ***Progress towards the clinic***
  - ➡ ***Application to tox testing***







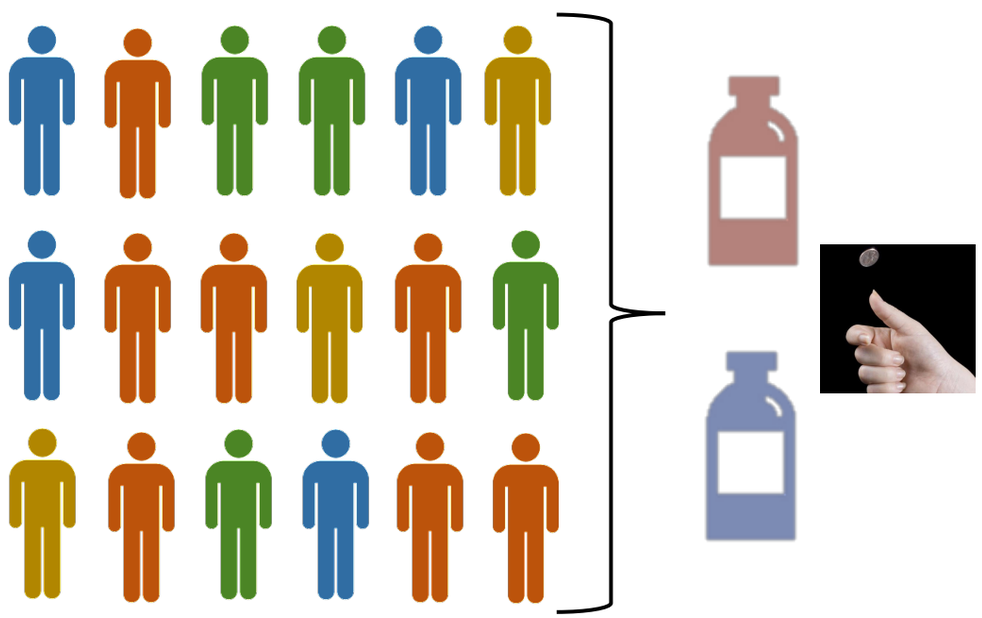




- ***Closing Thoughts***

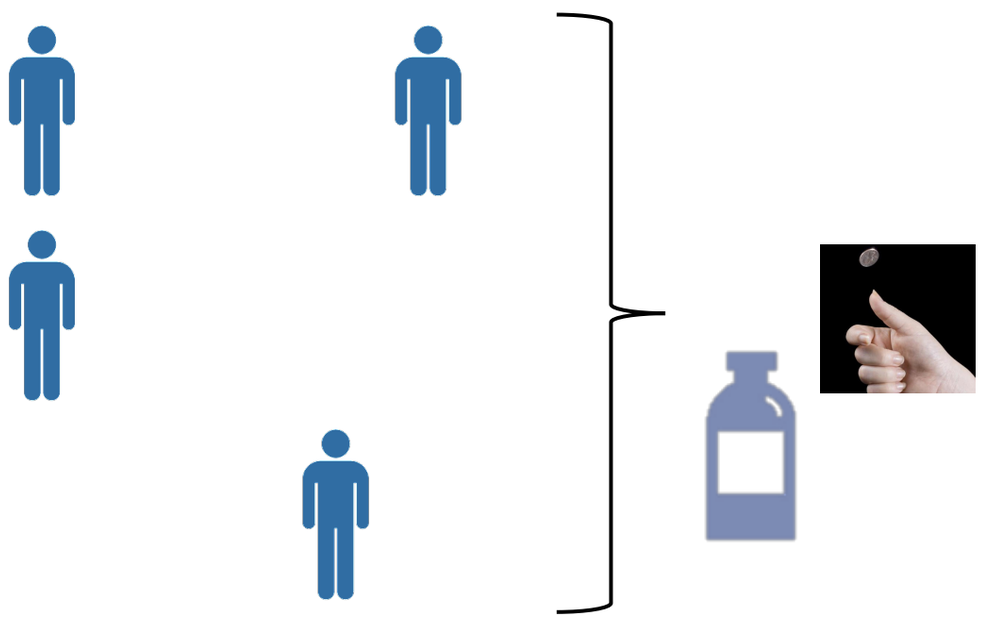
Cancer Treatment: Clinical Need

Current Paradigm

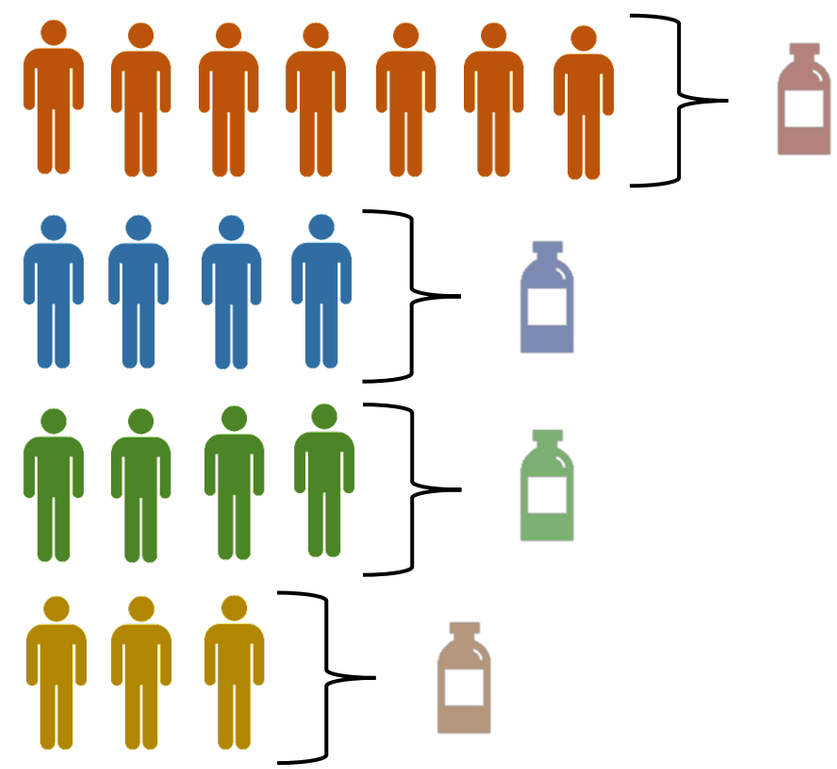


Cancer Treatment: Clinical Need

Current Paradigm

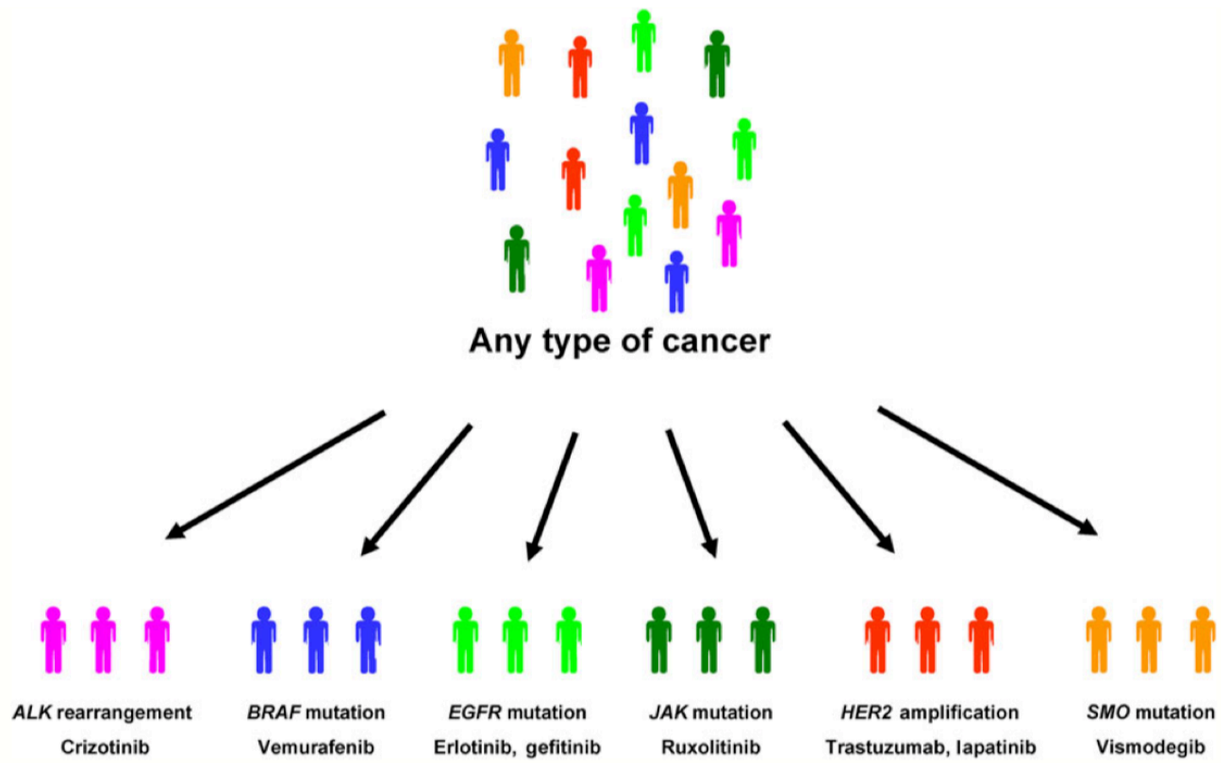


Goal



Genomics Alone is Not the Answer

Hype



Only ~10% are actionable

Reality

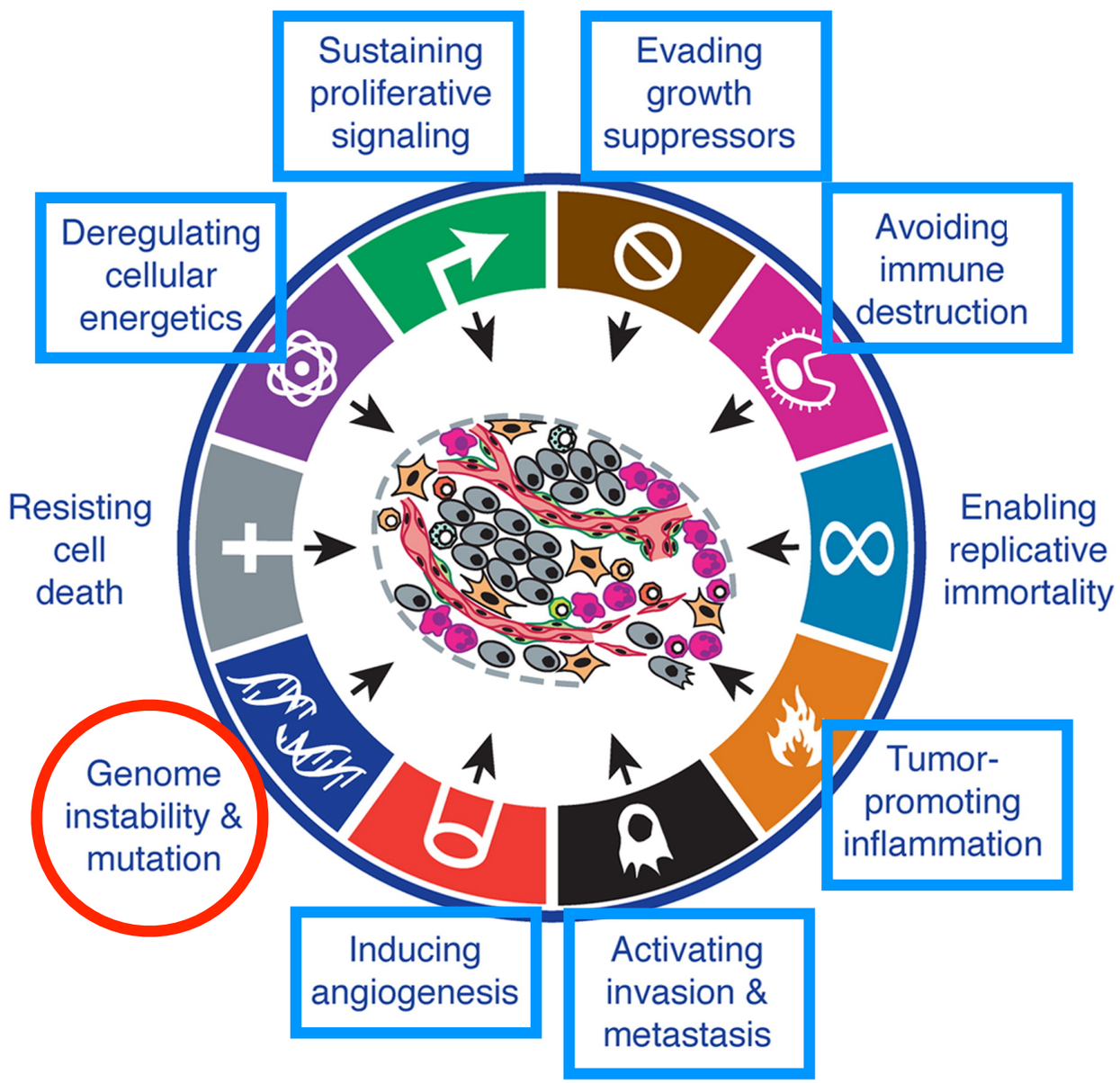
However, the majority of genomic alterations remain undefined with regard to their clinical significance

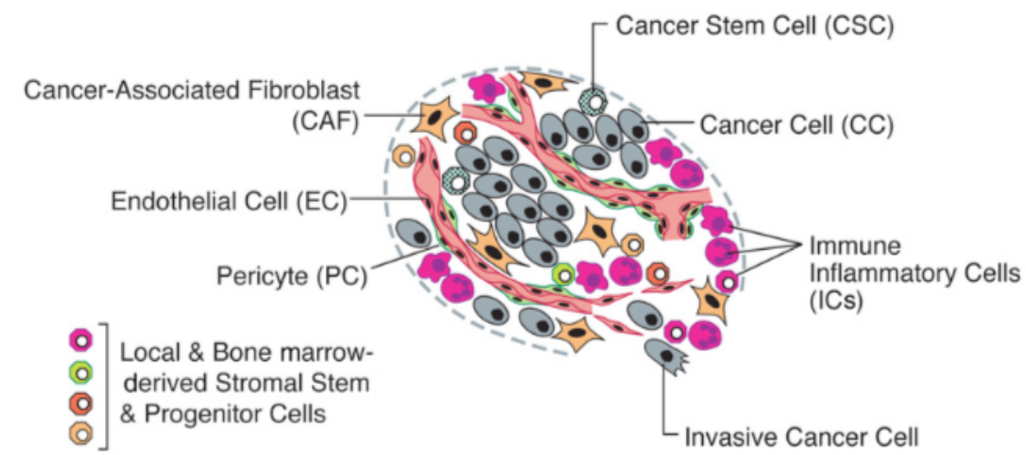
Most cancers become resistant to first line treatment in a few months

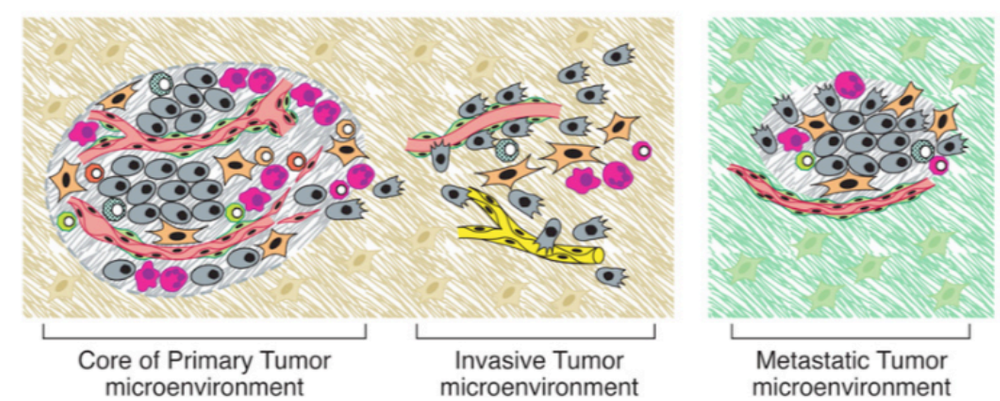
Kuo, 2016; Yuan, 2014; Dienstmann, 2015

Hallmarks of Cancer

Cancer is more than genetic mutations



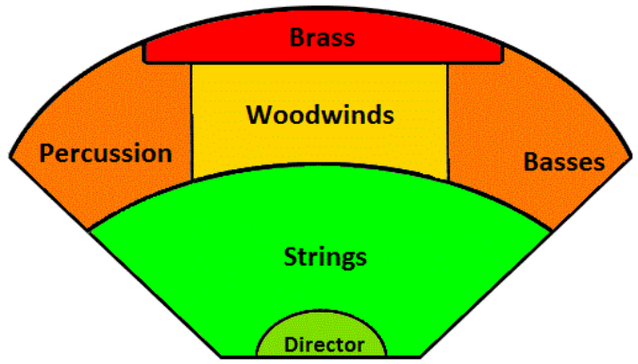


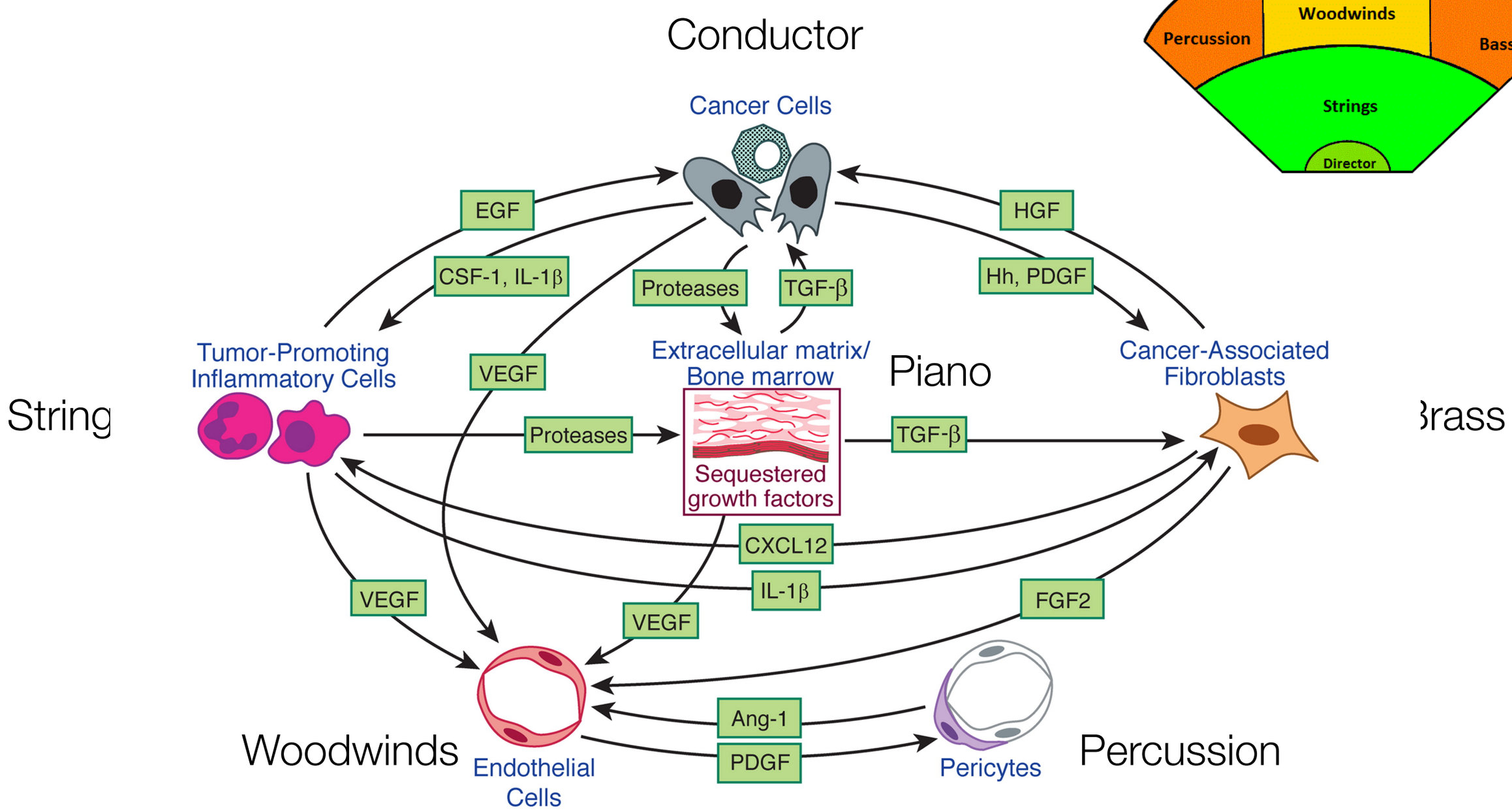


Hanahan & Weinberg, Cell 2011

Intercell Signaling

Need informed model development





Hanahan & Weinberg, Cell 2011

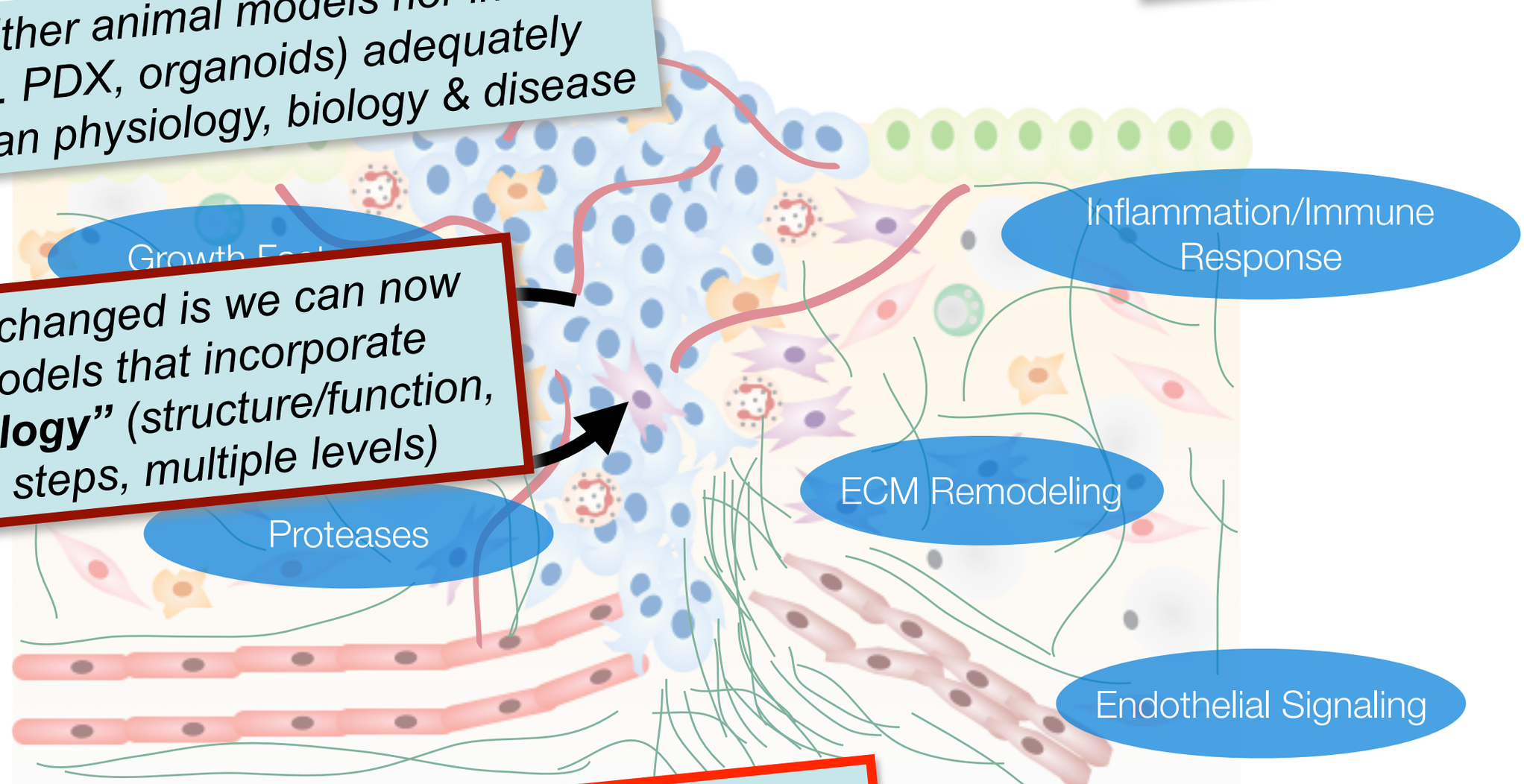
Complex Interactions Regulate Cancer Progression

Quantifying & manipulating these interactions ***in vivo*** is challenging

Can ***organotypic*** models help?

Beyond genomics

Dilemma - neither animal models nor in vitro/ ex vivo (e.g. PDX, organoids) adequately replicate human physiology, biology & disease



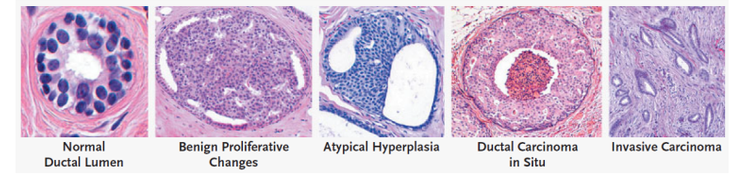
*What has changed is we can now make models that incorporate **"better biology"** (structure/function, multiple steps, multiple levels)*

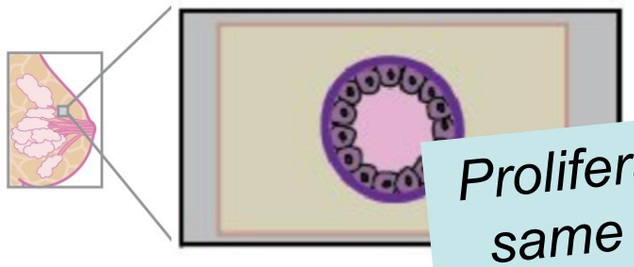
But how much is enough?

Can new models improve toxicity testing?

Clinically useful?

“Better” Biology Via Structure & Microenvironment (e.g. fibroblasts)

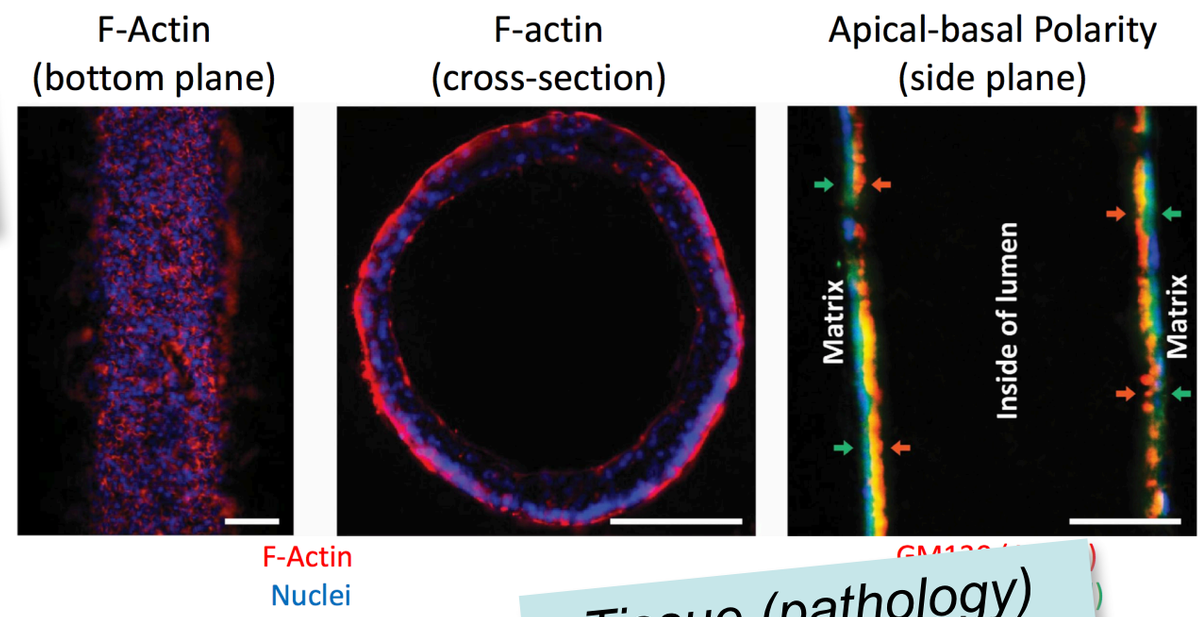




Proliferation rates same as in vivo

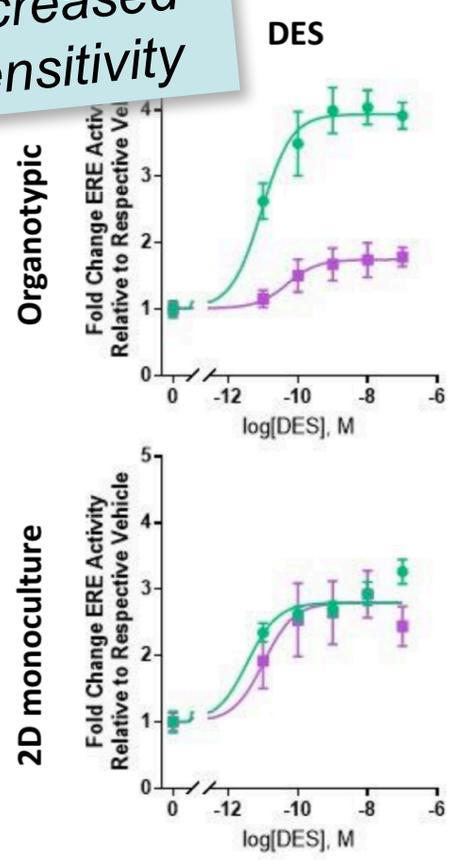
Proliferation

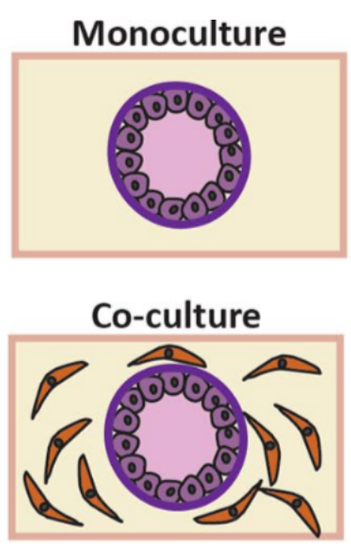
2D non-tumorigenic/tumorigenic	30-100%
3D duct.	**3**%
Normal in vivo	**3**%

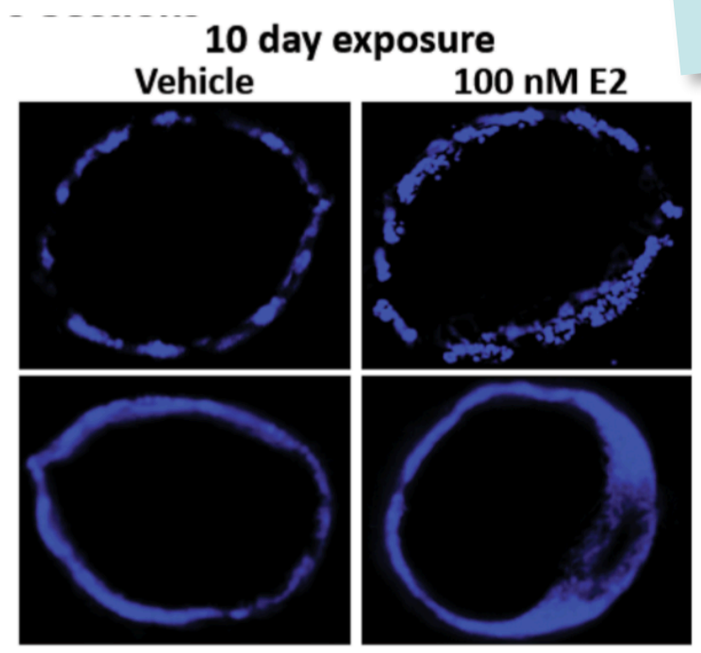


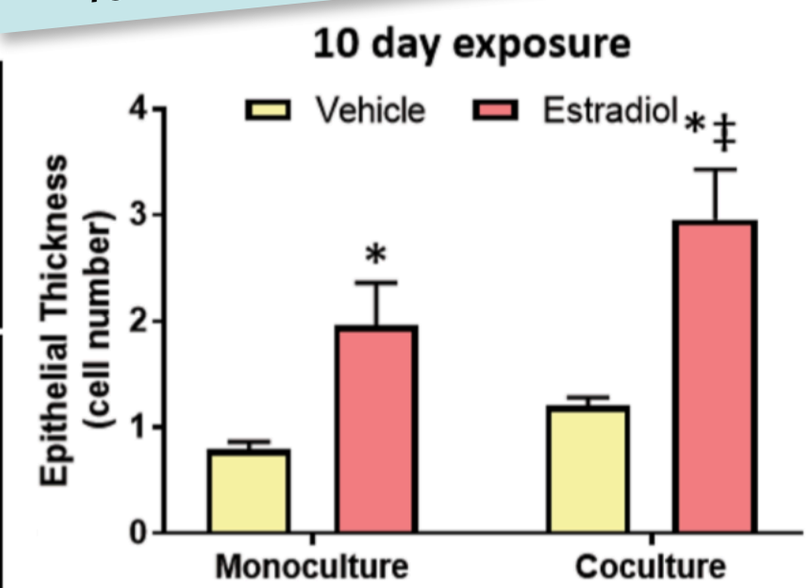
Tissue (pathology) level endpoints

Increased sensitivity





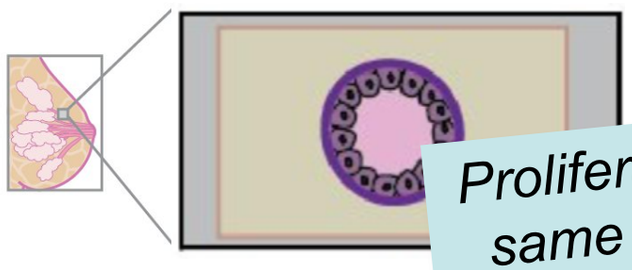




Morgan, et al., Scientific Reports, 2018

"Better" Biology Via Structure & Microenvironment (e.g. fibrob

Accumulating evidence of "better" biology

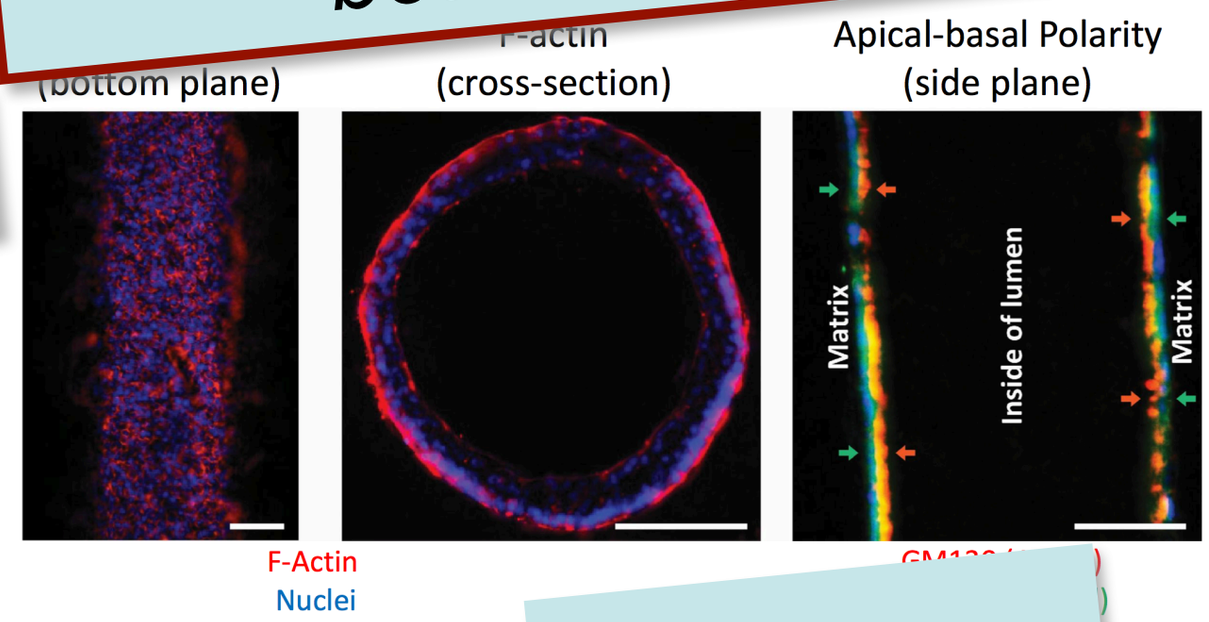


Proliferation rates same as in vivo

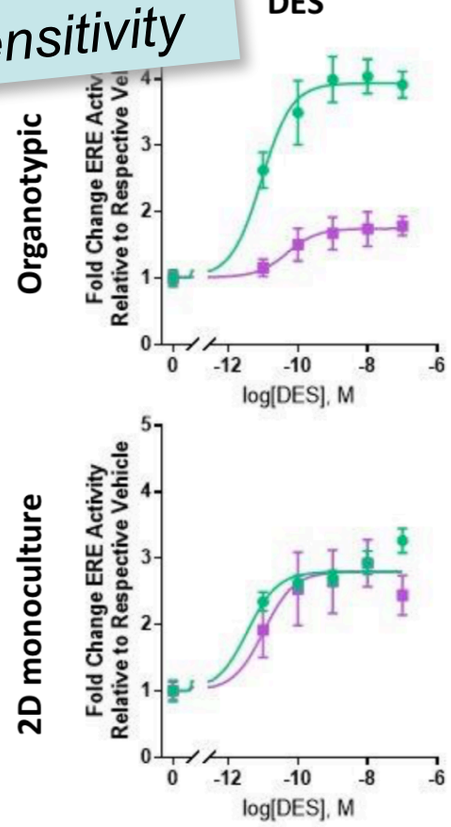
Proliferation

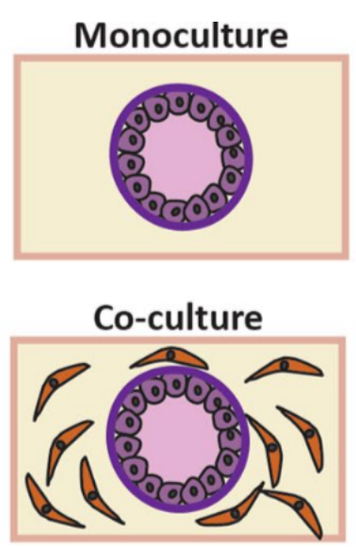
2D non-tumorigenic/tumorigenic	30-100%
3D duct.	**3**%
Normal in vivo	**3**%

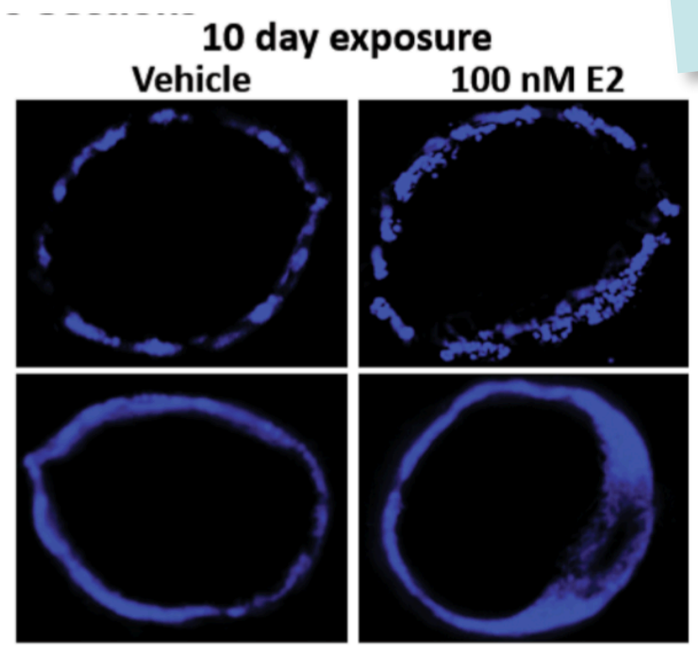
Increased sensitivity

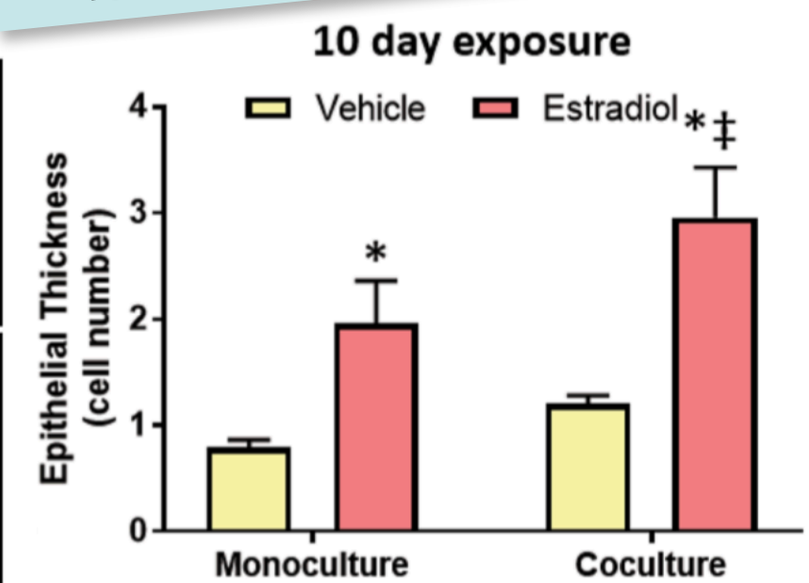


level endpoints







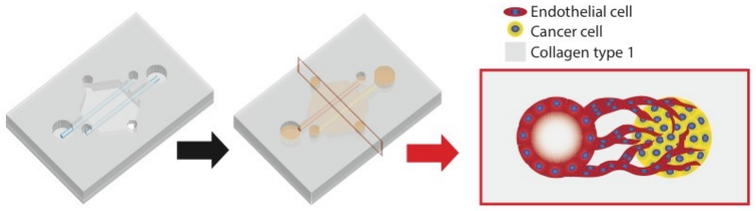


Morgan, et al., Scientific Reports, 2018

To The Clinic: Patient-specific Models to Predict Response

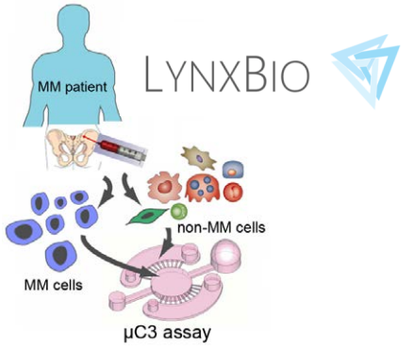
Renal Cell Carcinoma

Cancer-Vascular



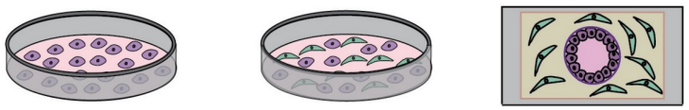
Multiple Myeloma

Cancer-Stromal



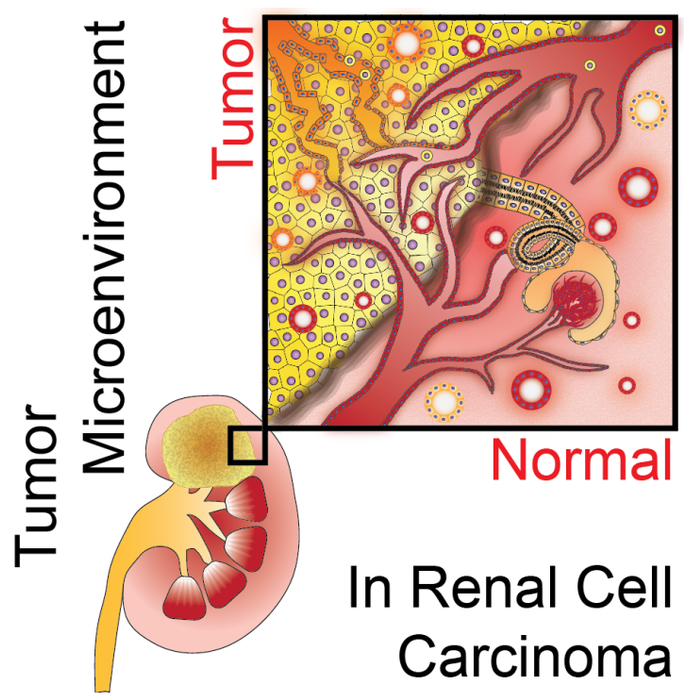
Breast Cancer

Tox Testing

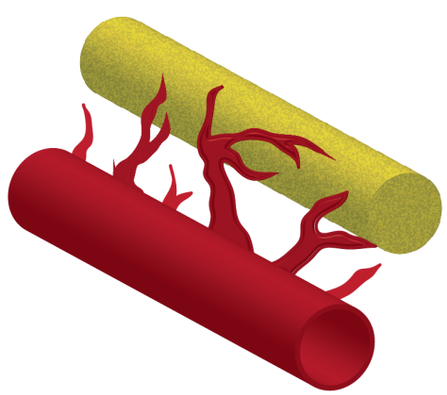


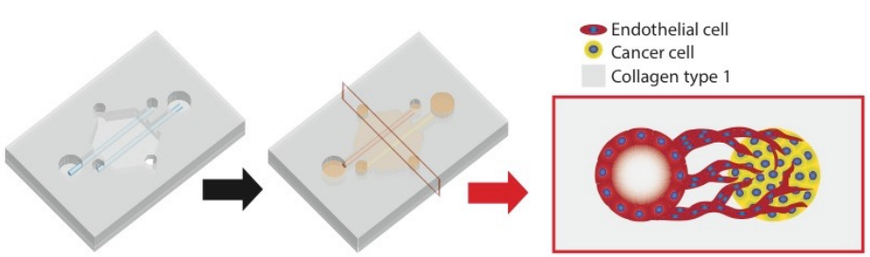
Cancer cell - endothelial interactions

Renal Cell Carcinoma



In Renal Cell Carcinoma





ARTICLE IN PRESS

EBIOM-02019; No of Pages 12

EBioMedicine xxx (2019) xxx

Contents lists available at ScienceDirect

EBioMedicine

journal homepage: www.ebiomedicine.com

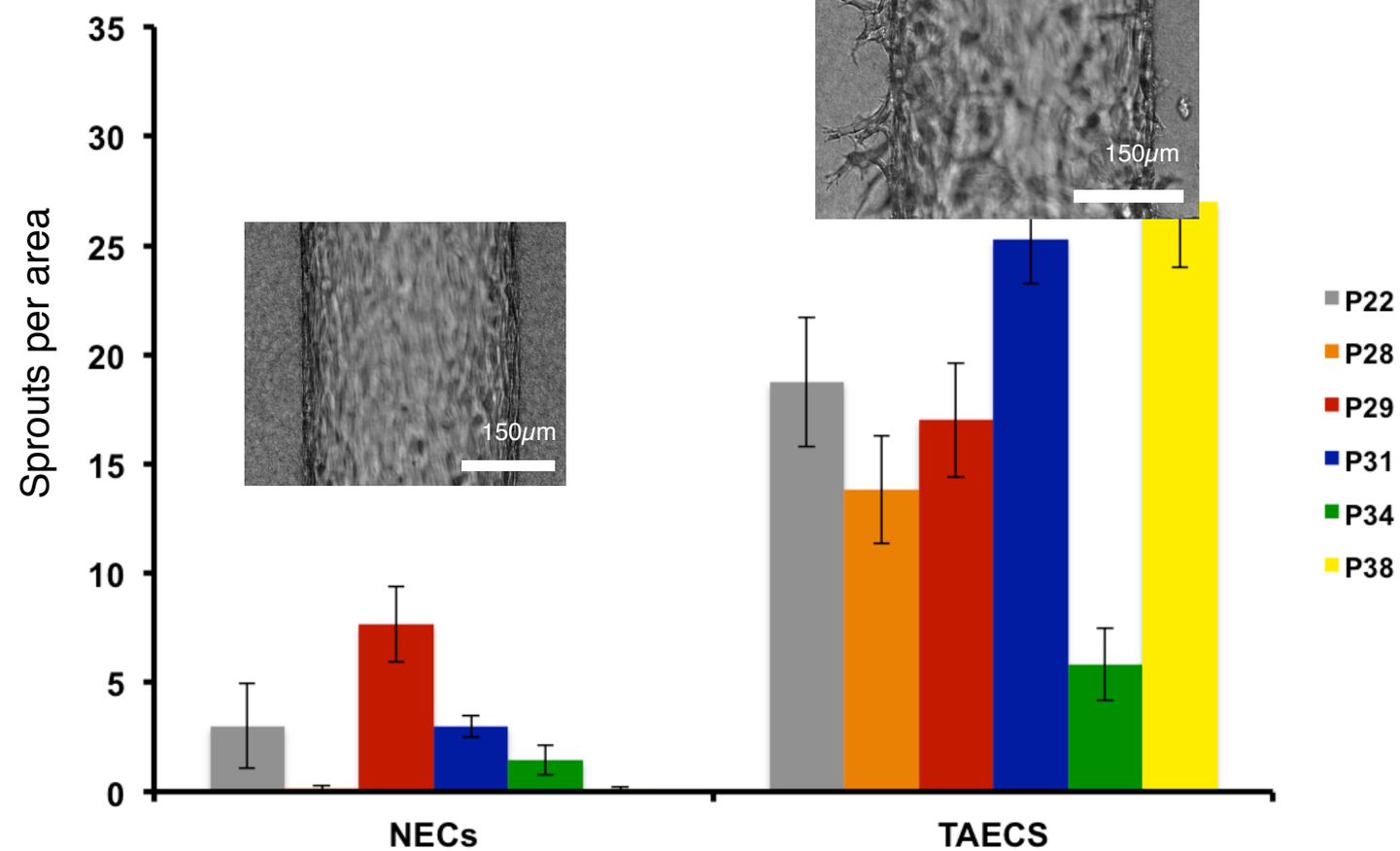
ELSEVIER

EBioMedicine
Published by THE LANCET

Patient-specific organotypic blood vessels as an *in vitro* model for anti-angiogenic drug response testing in renal cell carcinoma

José A. Jiménez-Torres [a,b,1], María Virumbrales-Muñoz [a,b,1], Kyung E. Sung [c], Moon Hee Lee [d], E. Jason Abel [d], David J. Beebe [a,b,e,*]

Endothelial characterization



Tony Jimenez, Maria Virumbrales-Munoz w/Jason Abel

Cancer cell - endothelial interactions

Renal Cell Carcinoma

Tumor

Tumor benvironment

ARTICLE IN PRESS

EBIOM-02019; No of Pages 12

EBioMedicine xxx (2019) xxx

Contents lists available at ScienceDirect

EBioMedicine

journal homepage: www.ebiomedicine.com

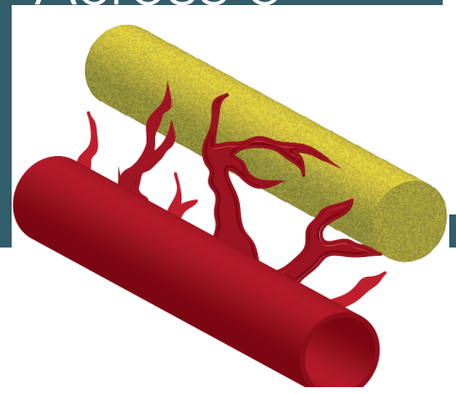
EBioMedicine Published by THE LANCET

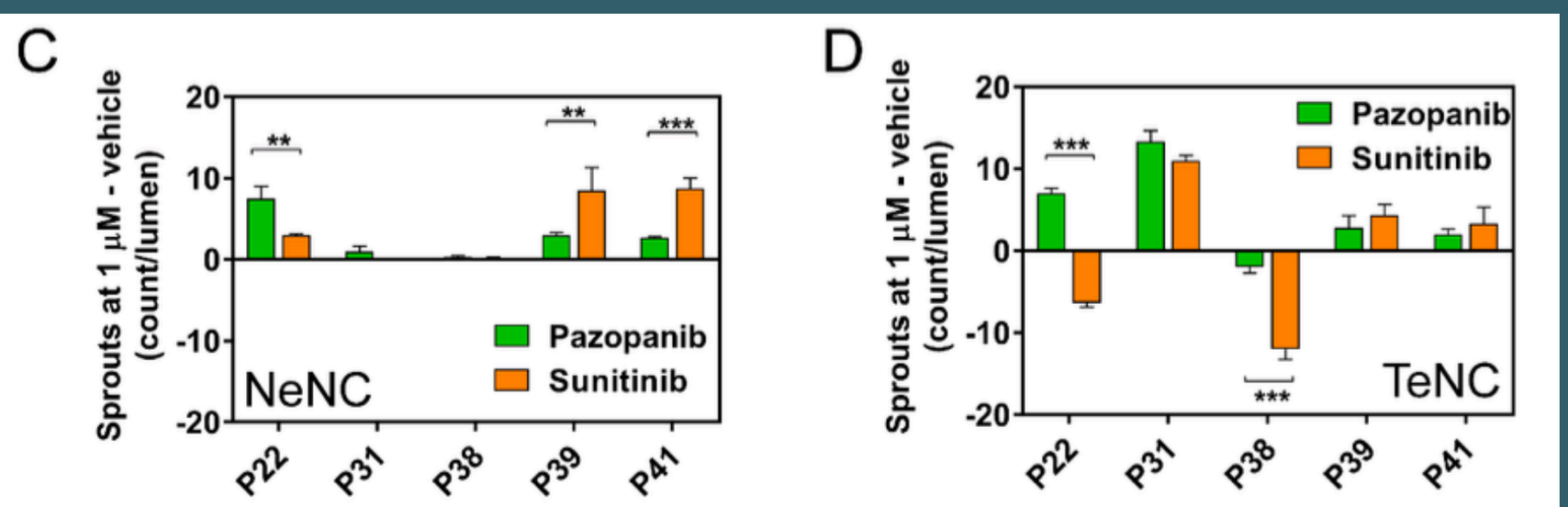
Patient-specific organotypic blood vessels as an *in vitro* model for anti-angiogenic drug response testing in renal cell carcinoma

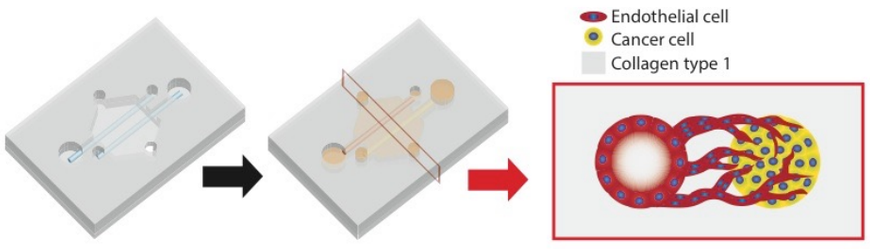
José A. Jiménez-Torres [a,b,1], María Virumbrales-Muñoz [a,b,1], Kyung E. Sung [c], Moon Hee Lee [d], E. Jason Abel [d], David J. Beebe [a,b,e,*]

Endothelial characterization

Differential Response Across 5







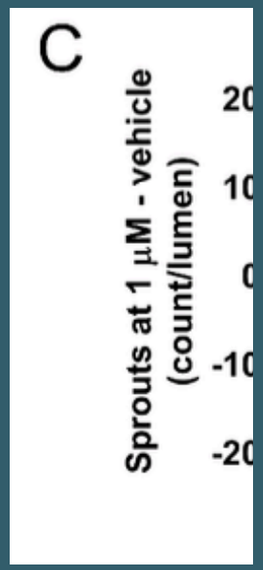
Tony Jimenez, Maria Virumbrales-Munoz w/Jason Abel

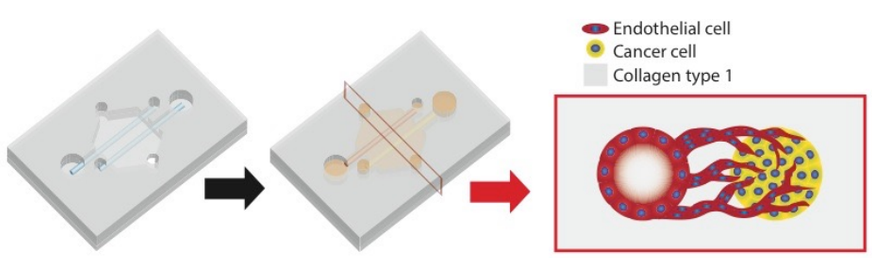
Renal Cell Carcinom

Tumor

Tumor environment

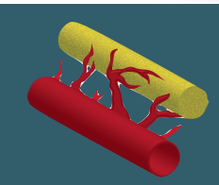
Differential Response Across 5 Patients



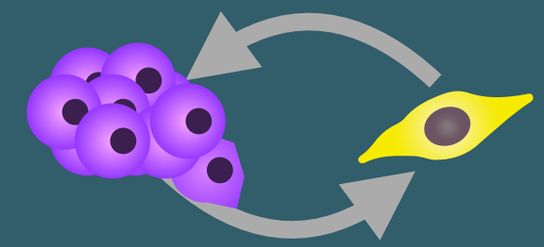


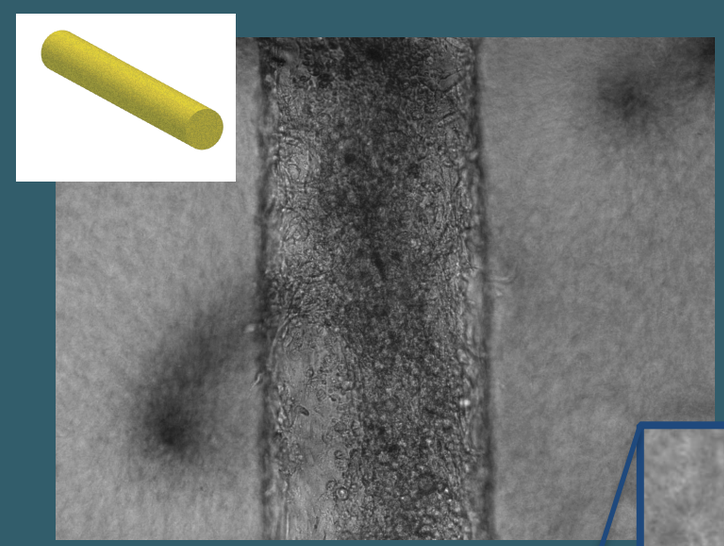
Cancer cell - endothelial interactions

Co-Culture (Epi/Endo)

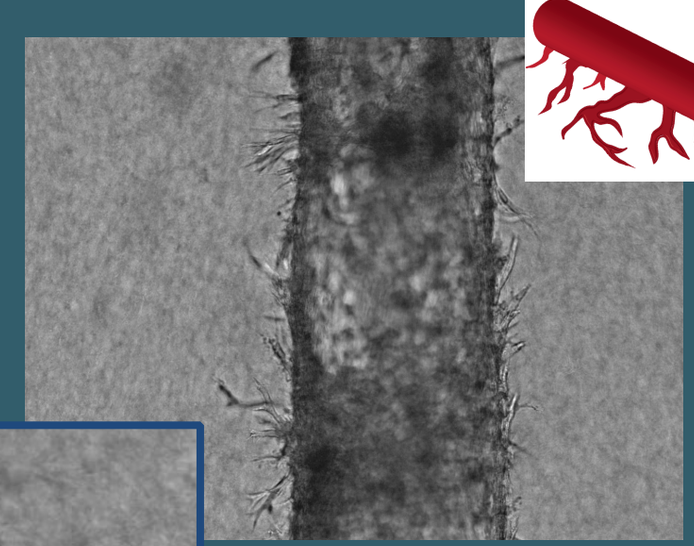


"conversation"

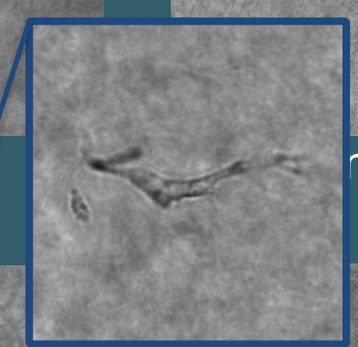


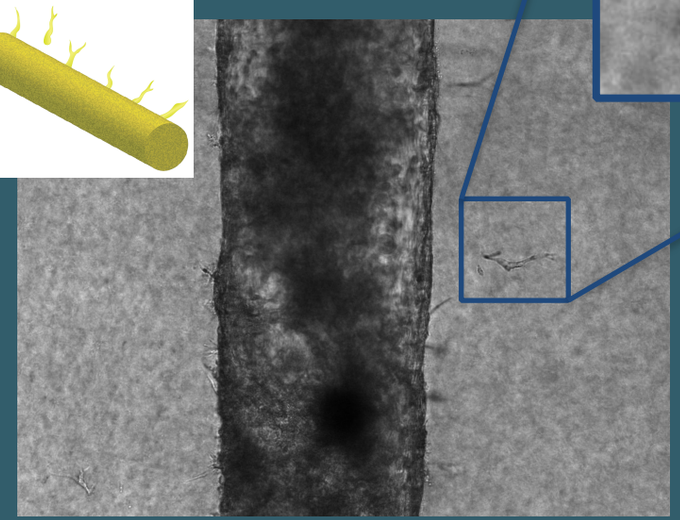


T29-3 Epi

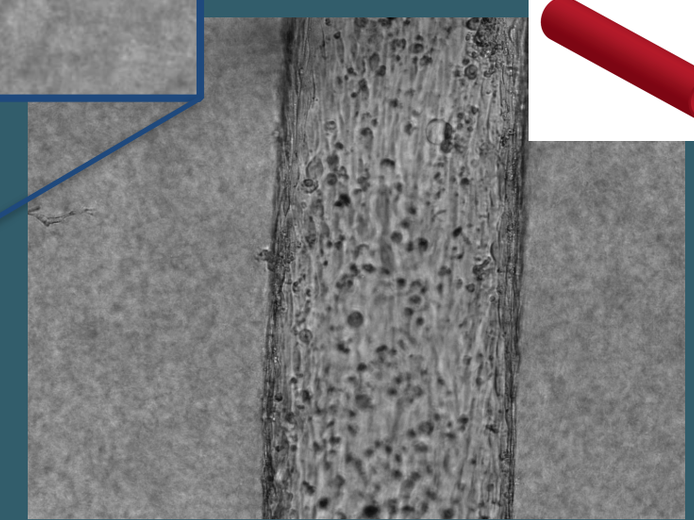


ndo





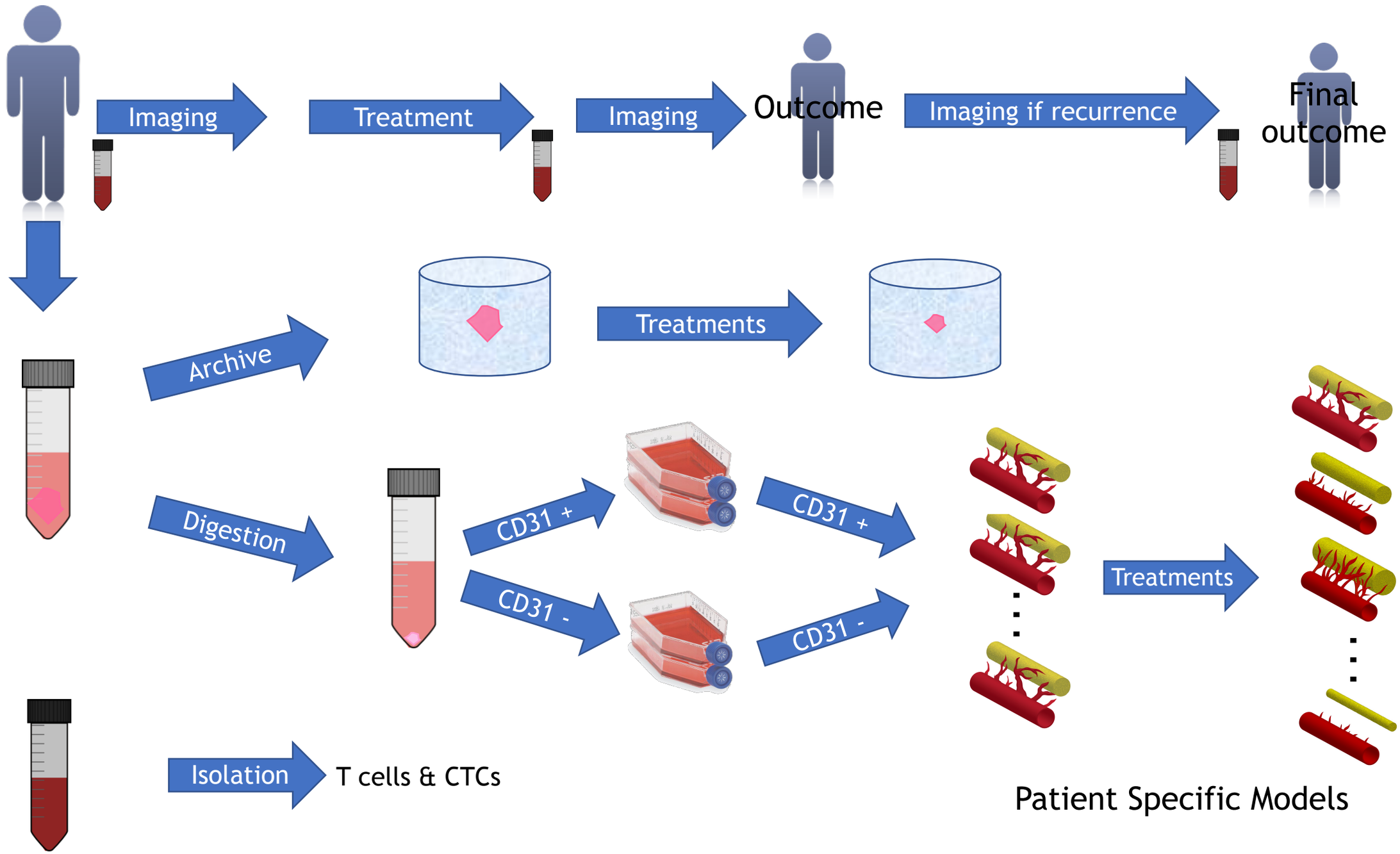
T29-3 Epi



T29-3 Endo

Clinical Study

NCI Moonshot Award

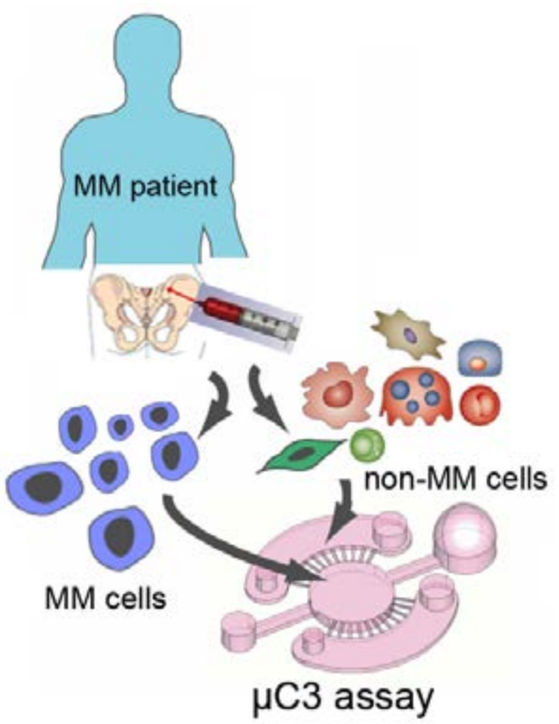


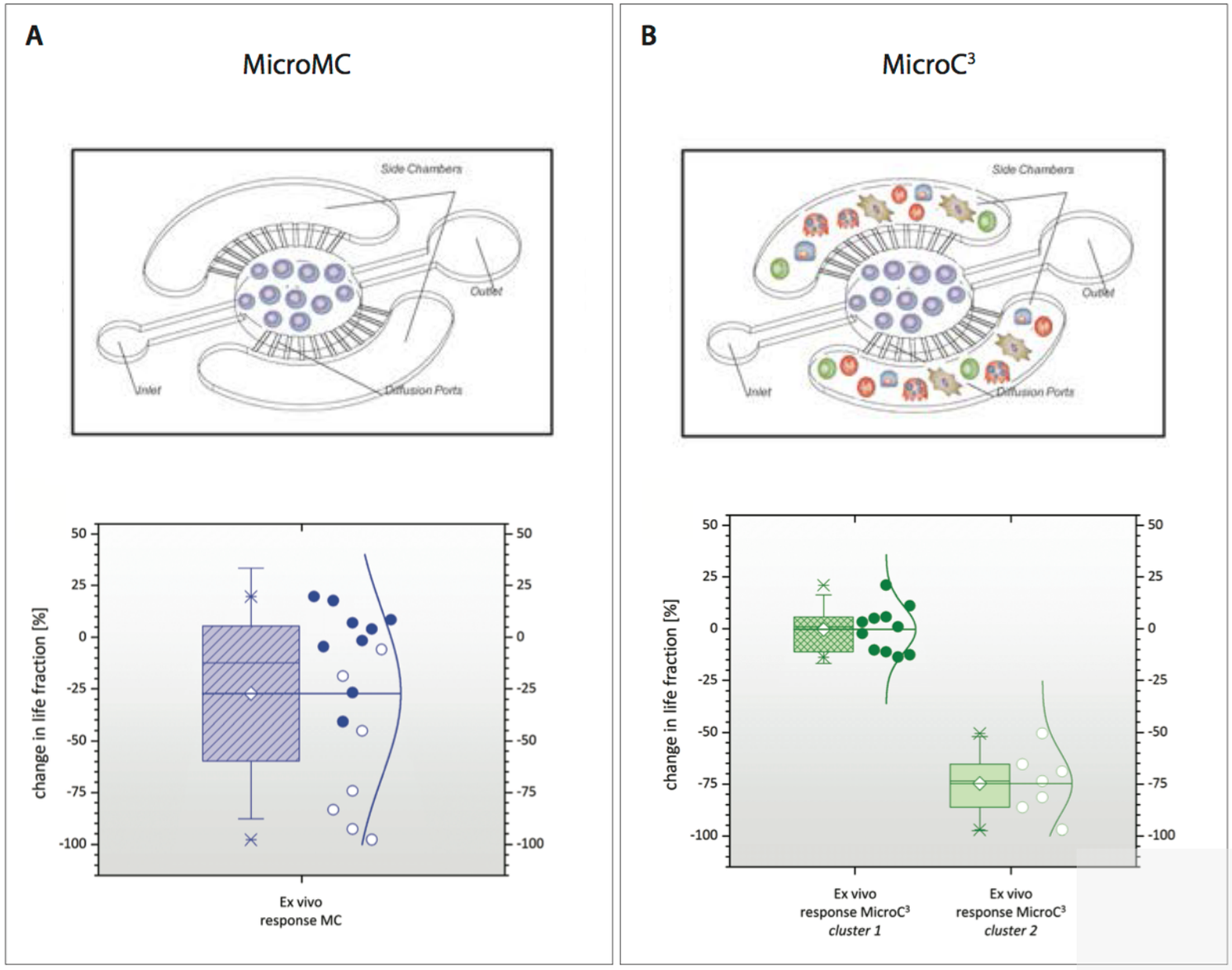
w/Jason Abel, Steve Cho, et al.

Predicting Patient Outcome

Small volumes = small #s of cells per endpoint

Enables testing with patient cells





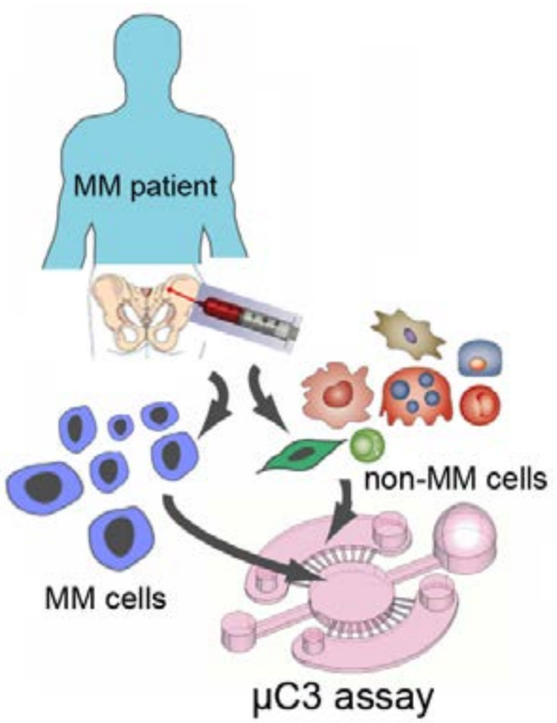
Young, Blood, 2012;
Pak, Integrative Biology, 2015

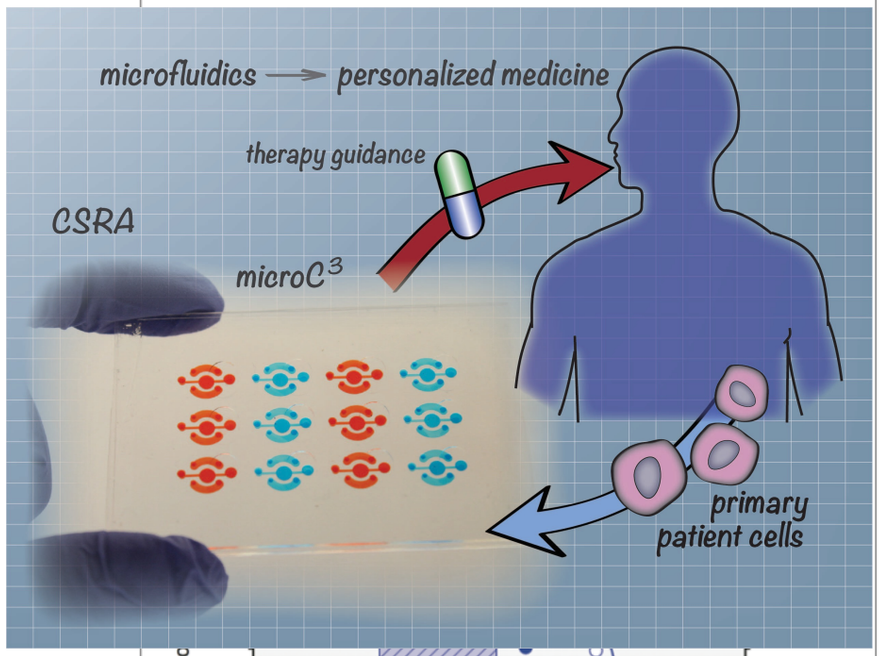
Sometimes not much is enough

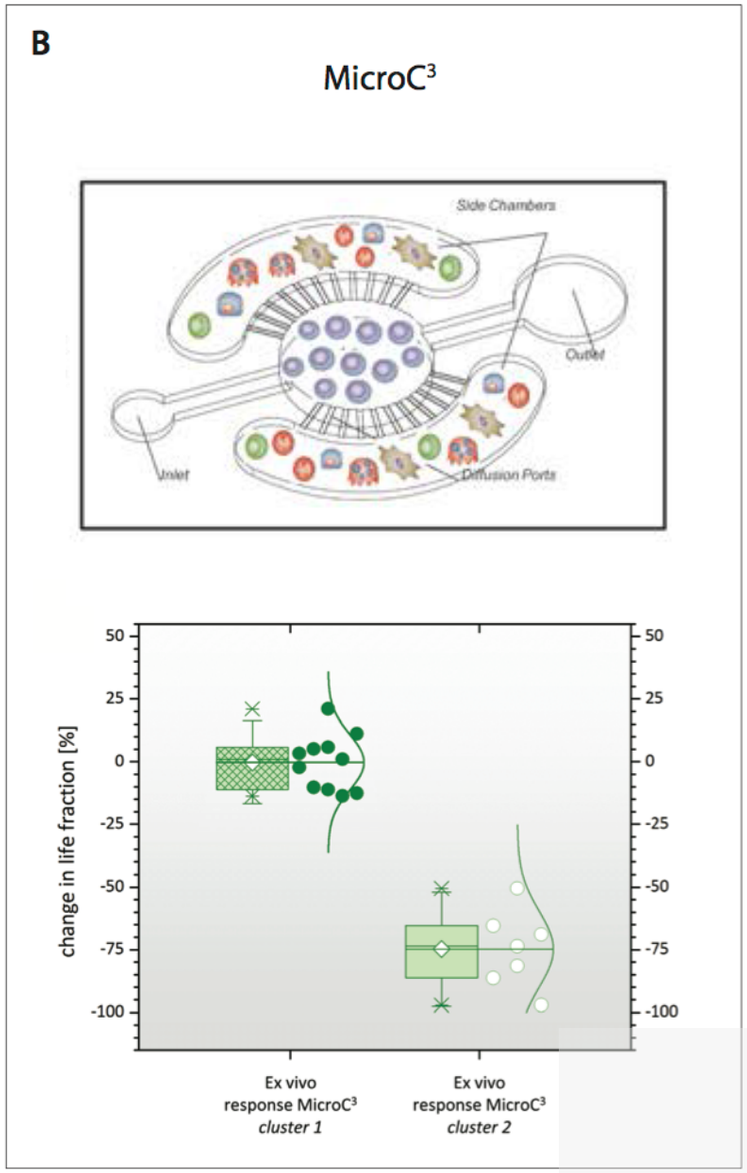
Predicting Patient Outcome

Small volumes = small #s of cells per endpoint

Enables testing with patient cells







Young, Blood, 2012;
Pak, Integrative Biology, 2015

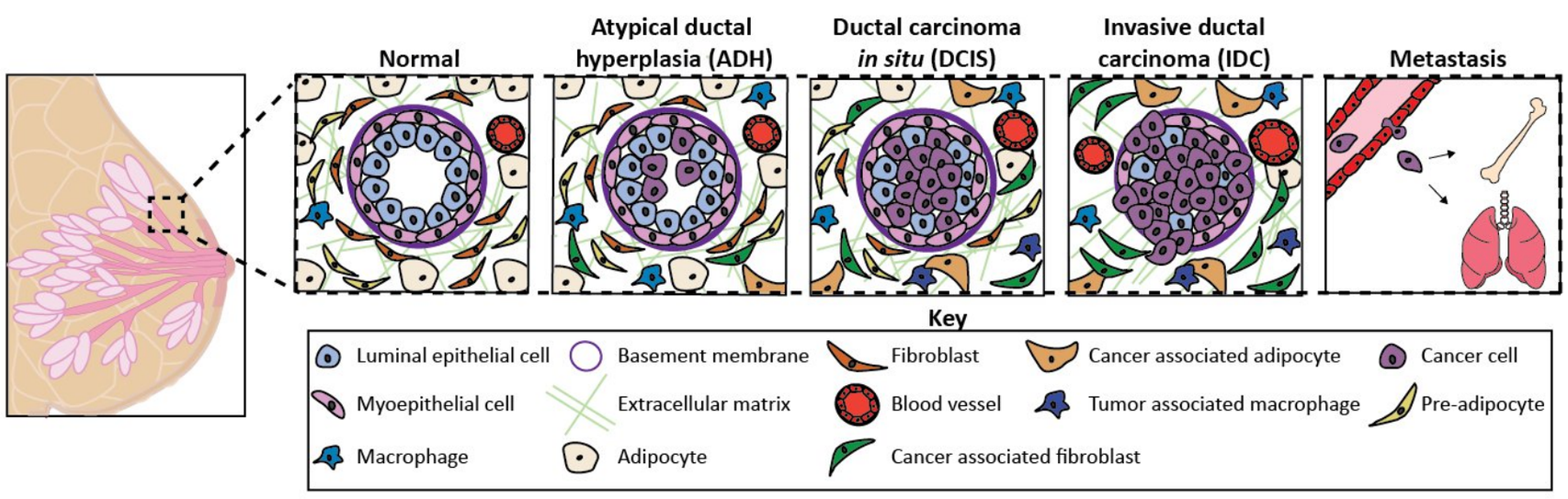
LYNXBIO

Application to Tox Testing

Do organotypic systems flag different chemical hits than a traditional *in vitro* platform?

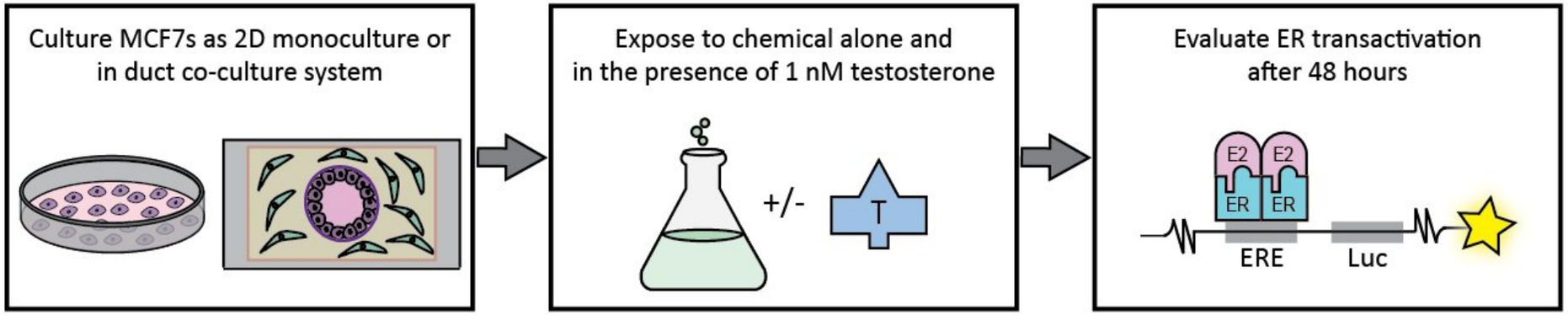
- **Overarching questions:** Does structure matter? How do mammary stromal cells regulate breast cancer cell responses to xenobiotics?

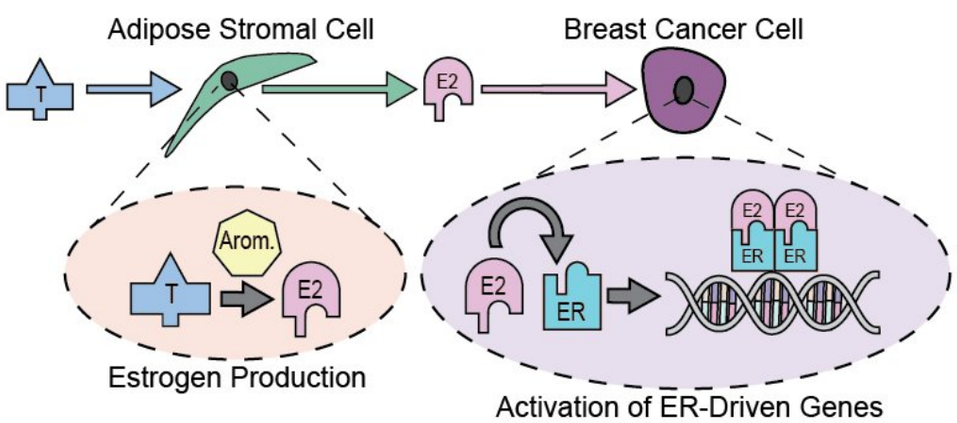
- **Premise:**
  - Estrogen receptor signaling is important to breast cancer progression
  - The microenvironment regulates the behavior and ER signaling of breast cancer cells, so may be important when testing chemicals *in vitro*
  - Our understanding of how chemicals affect breast cancer is **mostly from data collected using simple *in vitro* models (e.g. 2D)**



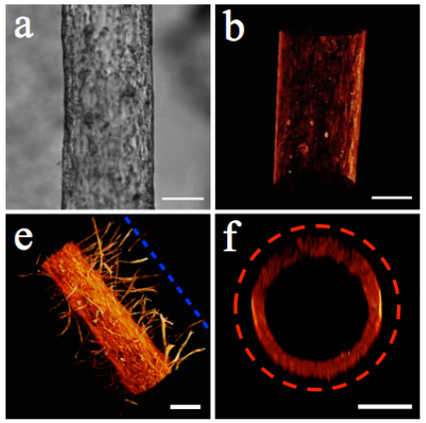
Application to Tox Testing

Approach





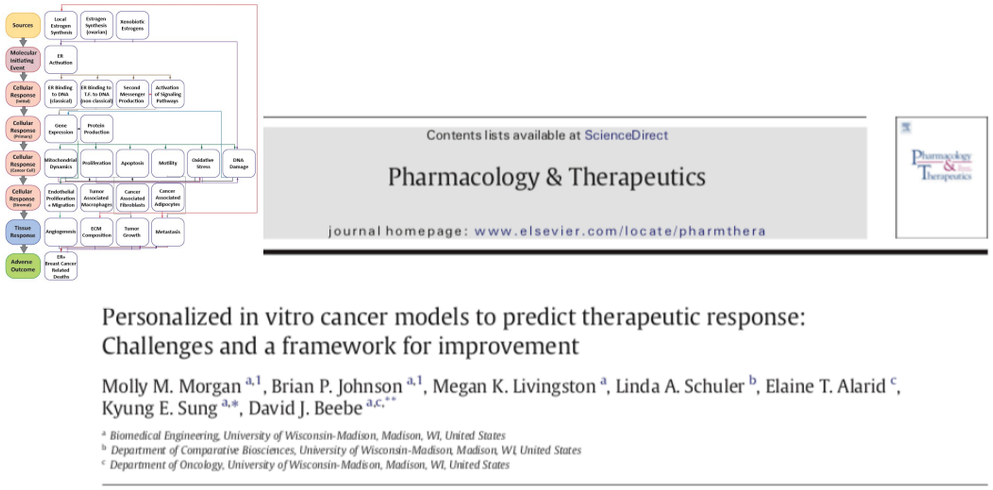
Mammary stromal cells indirectly regulate ER-driven responses by producing aromatase, the enzyme responsible for estrogen production



<u>How complex do we need to make *in vitro* models to predict *in vivo* responses?</u>

Methods/Approach

- MCF7-derived duct responds to E2 similar to already established *in vitro* and *in vivo* systems; responses mirror what is framed in AOP



Contents lists available at ScienceDirect

Pharmacology & Therapeutics

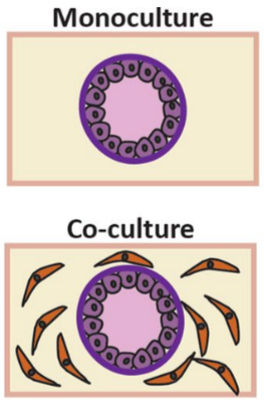
journal homepage: www.elsevier.com/locate/pharmthera

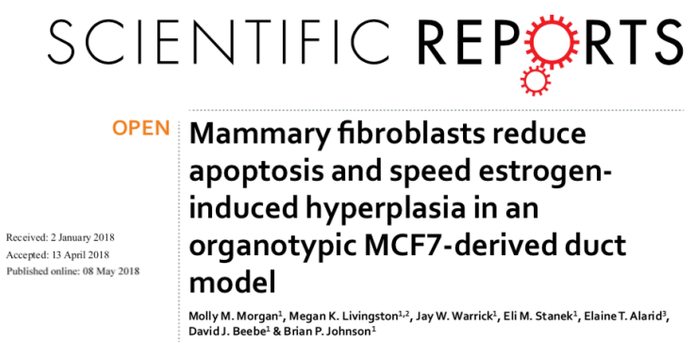
Personalized in vitro cancer models to predict therapeutic response: Challenges and a framework for improvement

Molly M. Morgan [a,1], Brian P. Johnson [a,1], Megan K. Livingston [a], Linda A. Schuler [b], Elaine T. Alarid [c], Kyung E. Sung [a,*], David J. Beebe [a,c,**]

[a] Biomedical Engineering, University of Wisconsin-Madison, Madison, WI, United States
[b] Department of Comparative Biosciences, University of Wisconsin-Madison, Madison, WI, United States
[c] Department of Oncology, University of Wisconsin-Madison, Madison, WI, United States

- When stromal cells are included in the matrix surrounding the breast duct, MCF7-derived ducts exhibit:
  - ***Molecular level:*** a reduction in ER protein, an increase in maximal ER transactivation
  - ***Cellular level:*** a decrease in apoptosis and increase in cell density
  - ***Tissue level:*** an increase in estrogen-induced hyperplasia





SCIENTIFIC REPORTS

OPEN

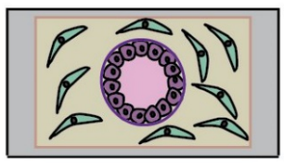
Mammary fibroblasts reduce apoptosis and speed estrogen-induced hyperplasia in an organotypic MCF7-derived duct model

Molly M. Morgan[1], Megan K. Livingston[1,2], Jay W. Warrick[1], Eli M. Stanek[1], Elaine T. Alarid[3], David J. Beebe[1] & Brian P. Johnson[1]

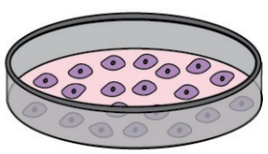
Received: 2 January 2018
Accepted: 13 April 2018
Published online: 08 May 2018

Chemical responses differ in MCF7-derived duct co-culture model

Organotypic

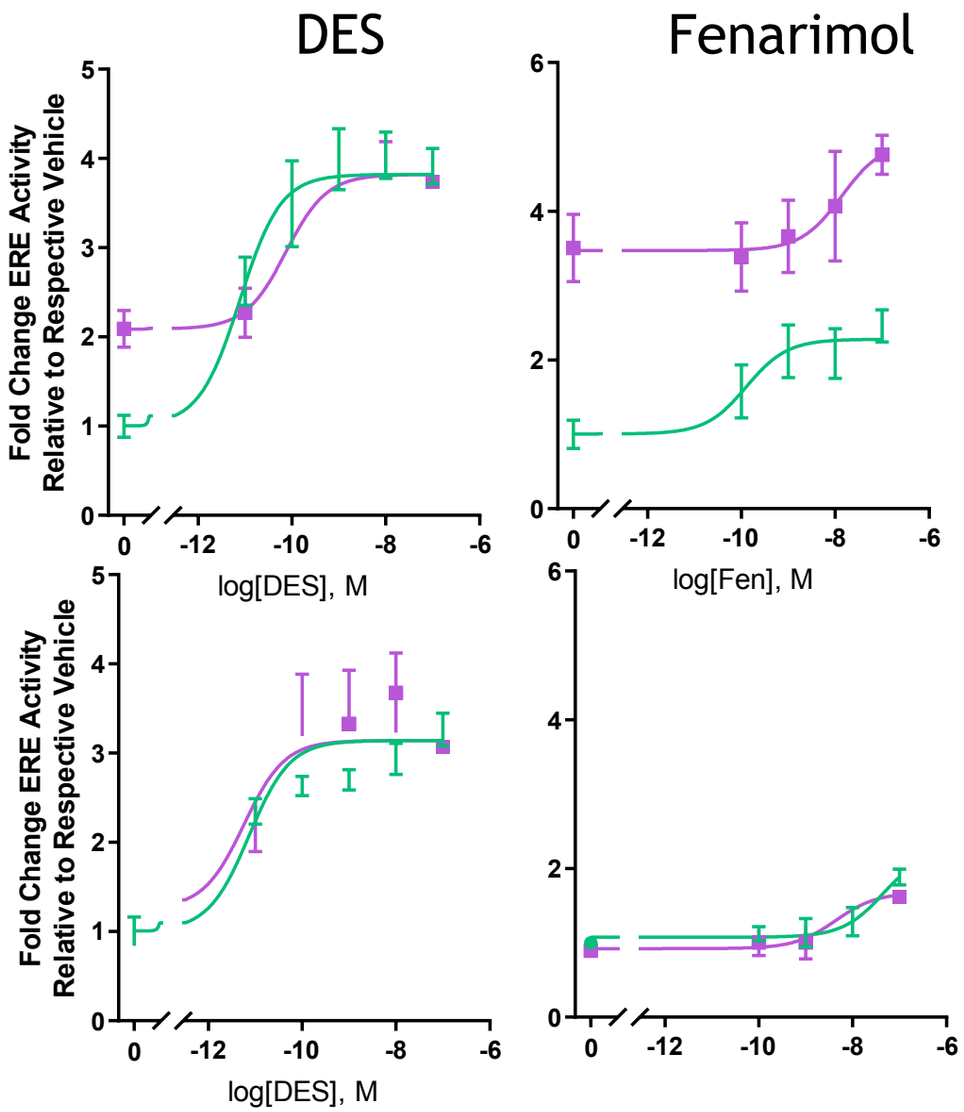


2D Monoculture

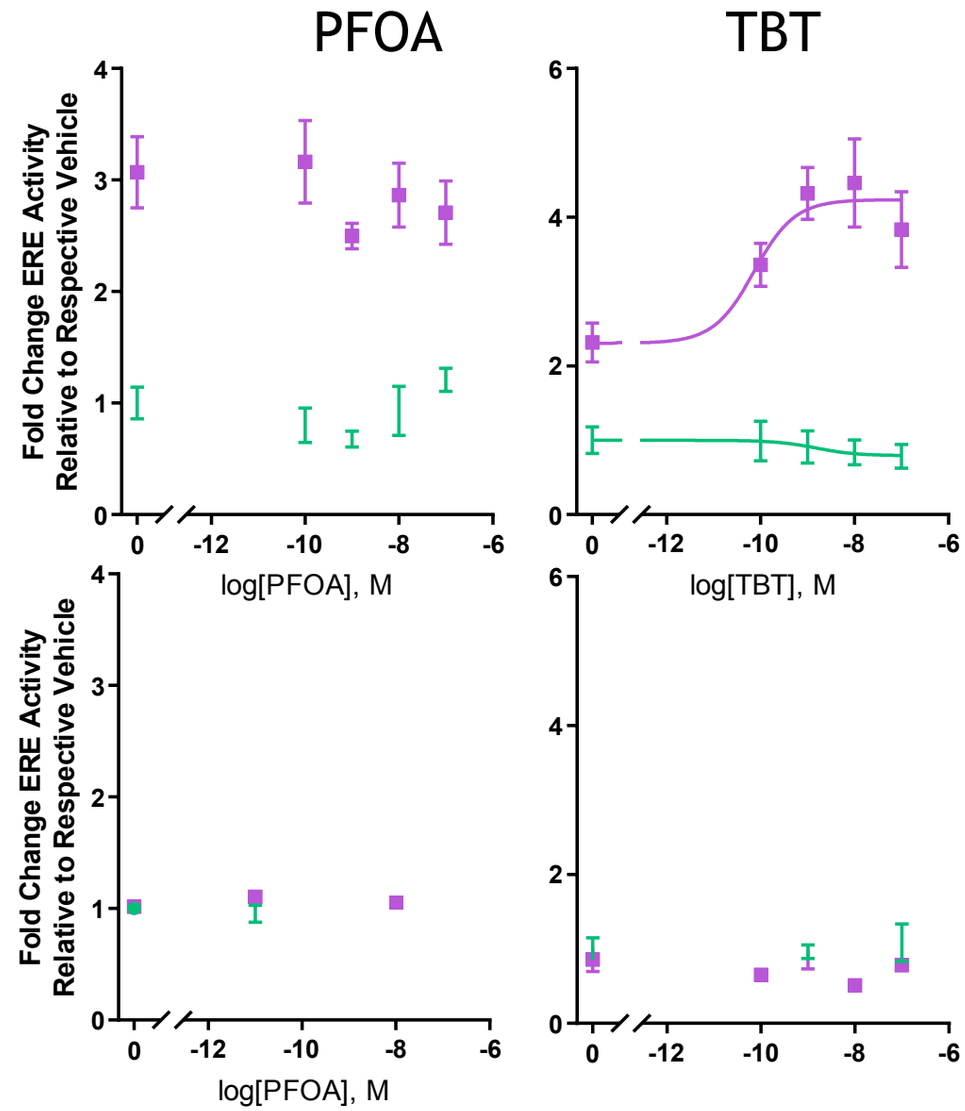


Without testosterone
With testosterone

Known ER Agonists



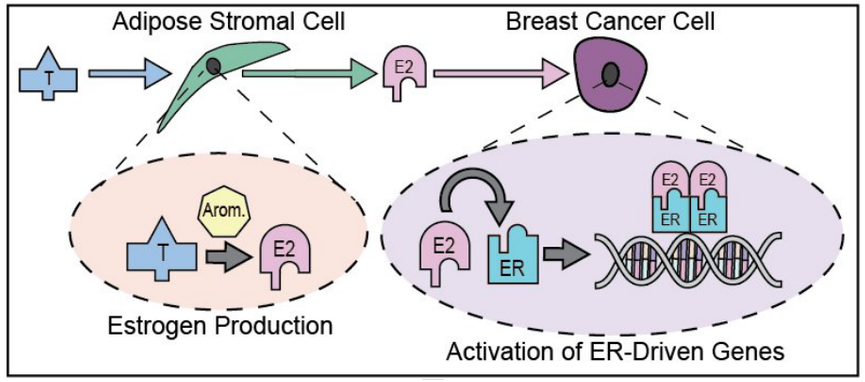
Uncertain ER activity

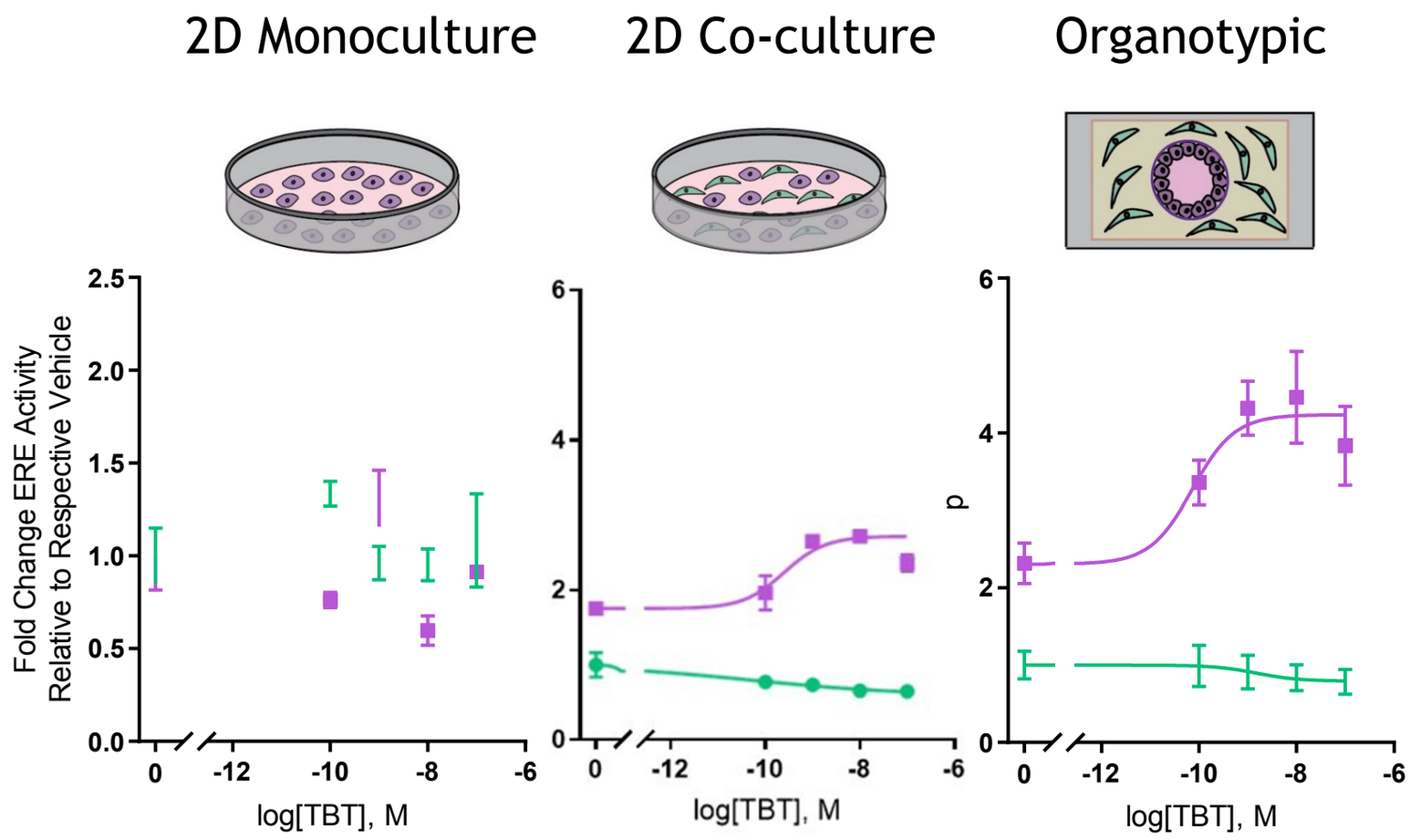


Organotypic model flags different chemical hits

But how much is enough?

Tributyltin chloride (TBT) induces ER transactivation in co-culture, but not monoculture breast model

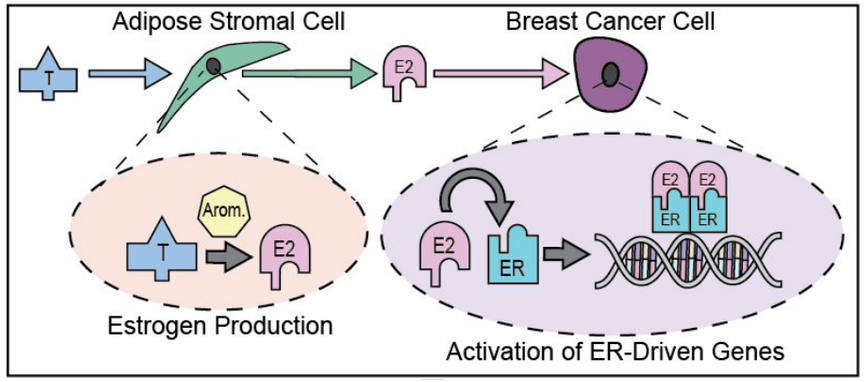




Some endocrine disrupting compounds, such as TBT, have complex effects that are missed when evaluated in a traditional 2D monoculture

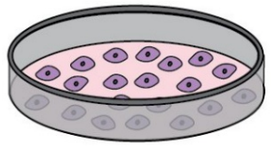
But how much is enough?

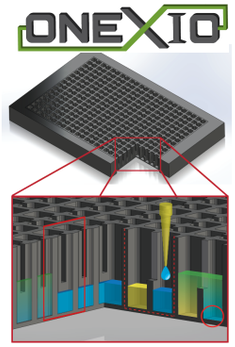
Tributyltin chloride (TBT) induces ER transactivation in co-culture, but not monoculture breast model



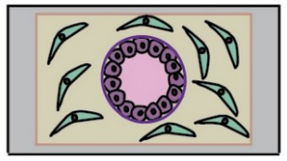
2D Co-culture

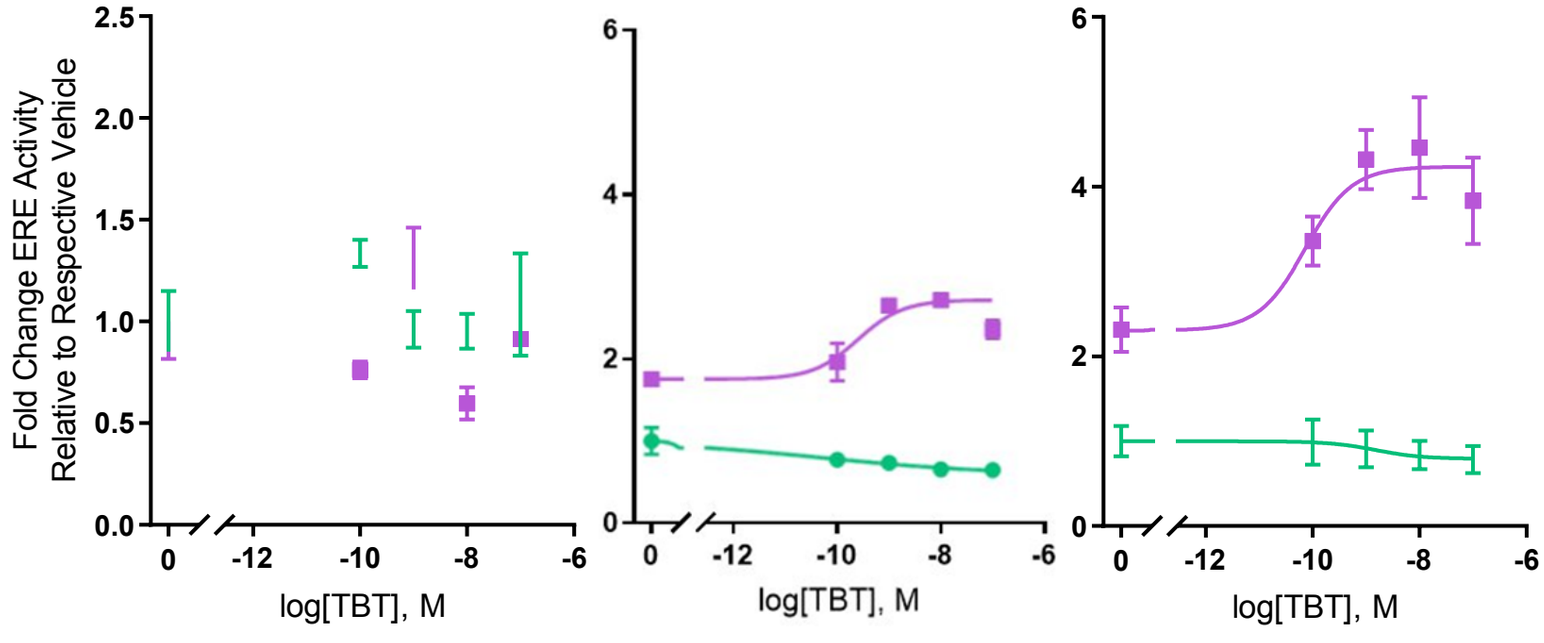
2D Monoculture





Organotypic





Some endocrine disrupting compounds, such as TBT, have complex effects that are missed when evaluated in a traditional 2D monoculture

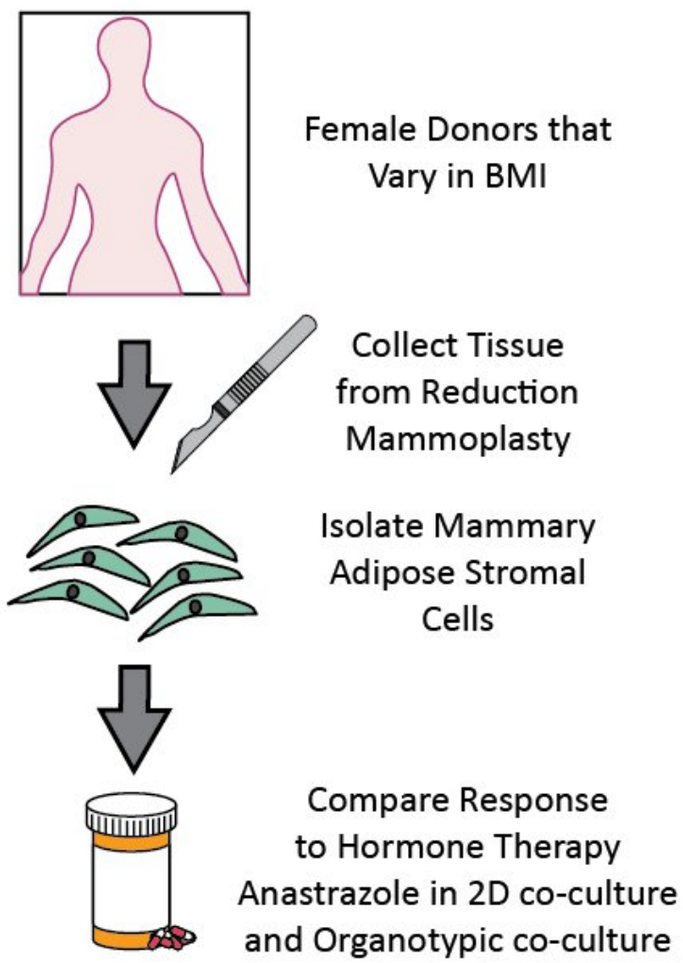
Without testosterone
With testosterone

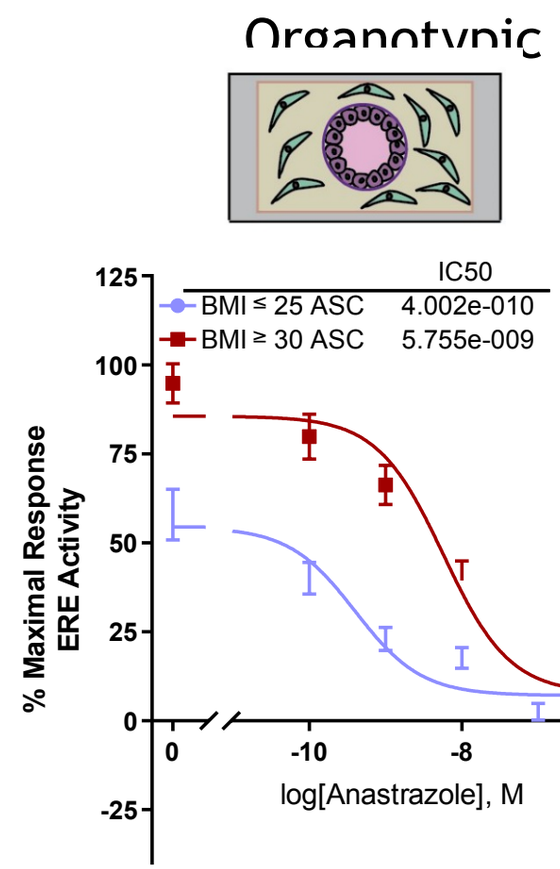
Organotypic, but not 2D culture models personal susceptibly differences to aromatase inhibition

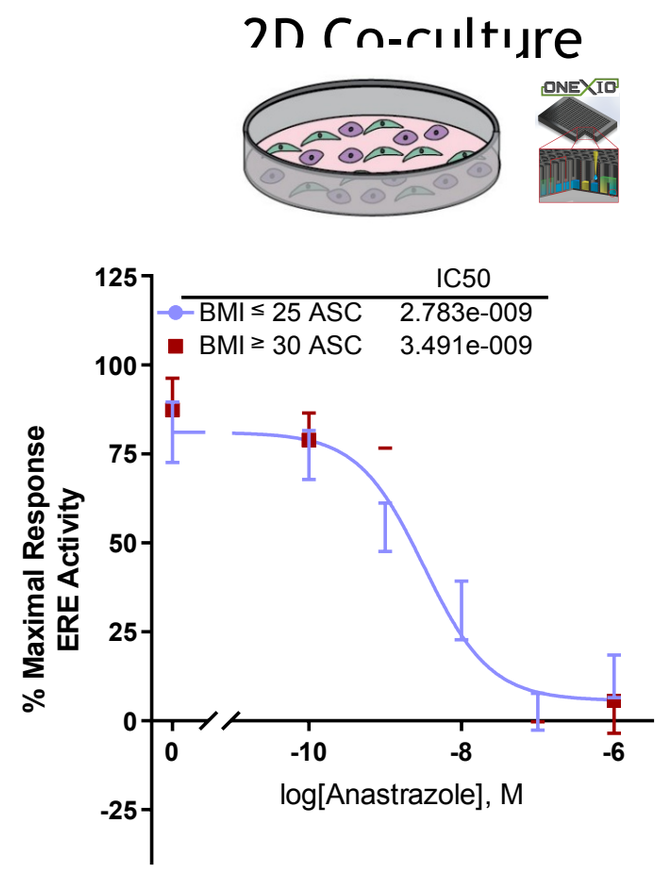
THE FASEB JOURNAL • RESEARCH • www.fasebj.org

Mammary adipose stromal cells derived from obese women reduce sensitivity to the aromatase inhibitor anastrazole in an organotypic breast model

Molly M. Morgan,* Lisa M. Arendt,† Elaine T. Alarid,‡ David J. Beebe,*,§,¶,1 and Brian P. Johnson§,2
*Department of Pathology and Laboratory Medicine, †Department of Comparative Biosciences, ‡Department of Oncology, §Department of Biomedical Engineering, and ¶Carbone Cancer Center, University of Wisconsin-Madison, Madison, Wisconsin, USA



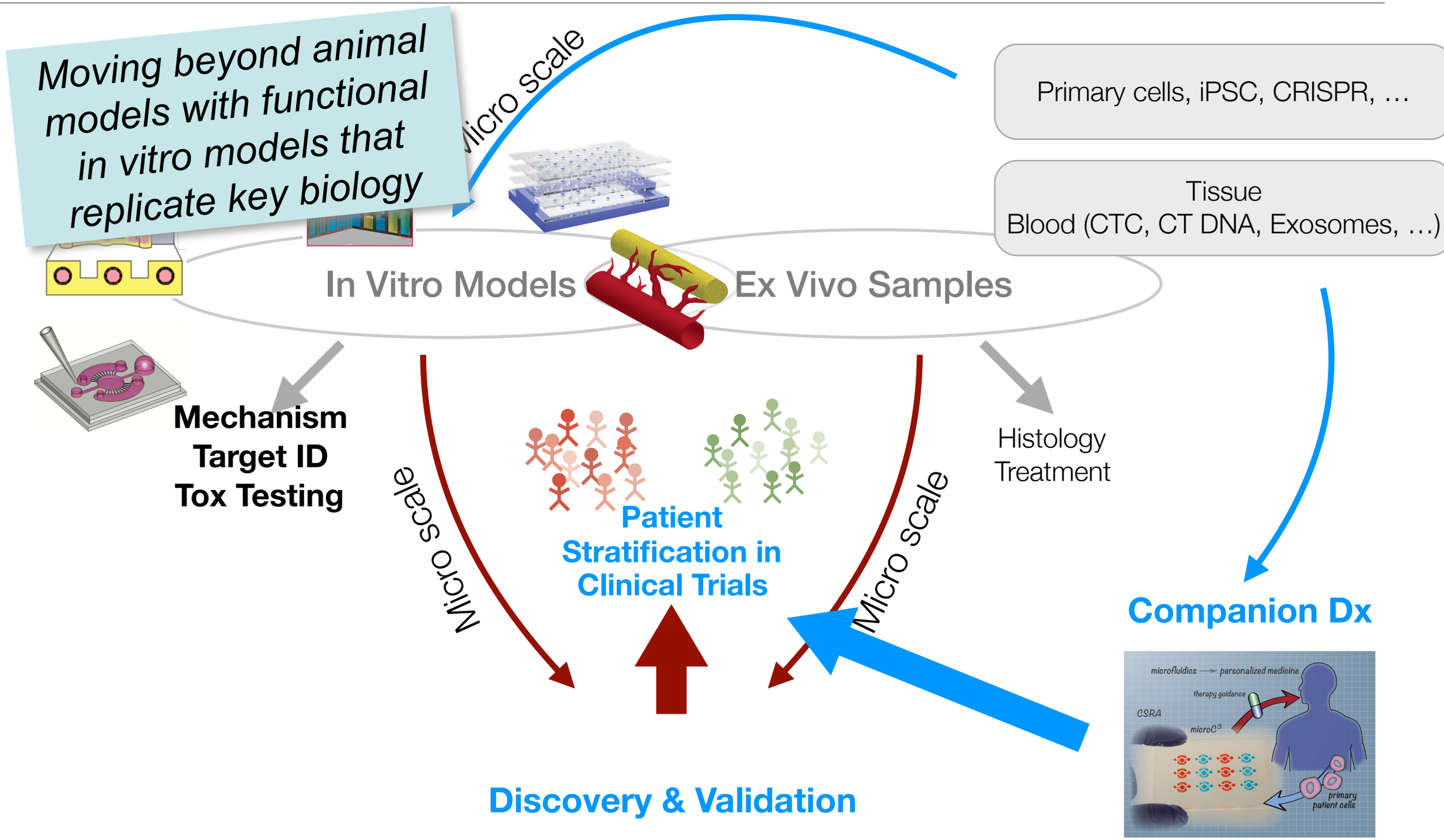




Chemical-induced aromatase inhibition differs in lean and obese individuals

Structure appears important

Improving Drug Discovery, Tox Testing & Clinical Outcomes



Improving Drug Discovery, Tox Testing & Clinical Outcomes

Moving beyond animal models with functional in vitro models that replicate key biology

Micro scale

Primary cells, iPSC, CRISPR, …

Tissue
Blood (CTC, CT DNA, Exosomes, …)

In Vitro Models

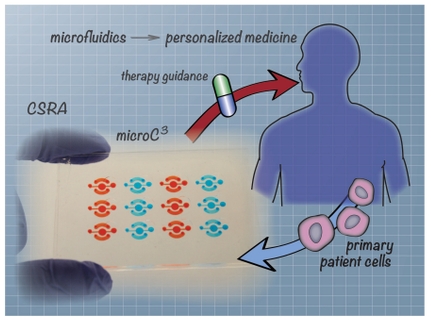
Ex Vivo Samples

Histology
Treatment

Micro scale

nt
tion in
Trials

Companion Dx



alidation

Improved models enable…

Repeated dosing

Tox testing

Biomarker development

Immunogenicity testing

Multi-target drug development

Target ID

And more…

Thanks to the Patients

Funding: NIH, DOD, PCF, EPA, MIR

mmb LAB

Collaborators: Andreas Friedl, Josh Lang, Elaine Alarid, Steve Cho, Anna Huttenlocher, Shigeki Miyamoto, Jason Abel, Hirak Basu, Will Ricke, Mark Burkard, Nancy Keller, Christina Hull, Richie Davidson, Melissa Rosenkranz, Melissa Skala, Dave Jarrard, Wie Huang, Lee Wilke, Christos Kyriakopoulos, …