Connecting Tissues and Investigators: Fibrosis in Health and Disease

December 6, 2017

Boston University Office of the Vice President and Associate Provost for Research

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Can We Reduce Fibrosis by Increasing the Levels of the Klotho Hormone?

Carmela Abraham

Professor
Biochemistry and Pharmacology &
Experimental Therapeutics, MED

Clotho, Zeus' daughter who spins the thread of life. Klotho gene named after her by Dr. Kuro-o

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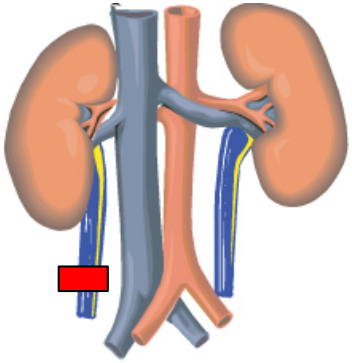
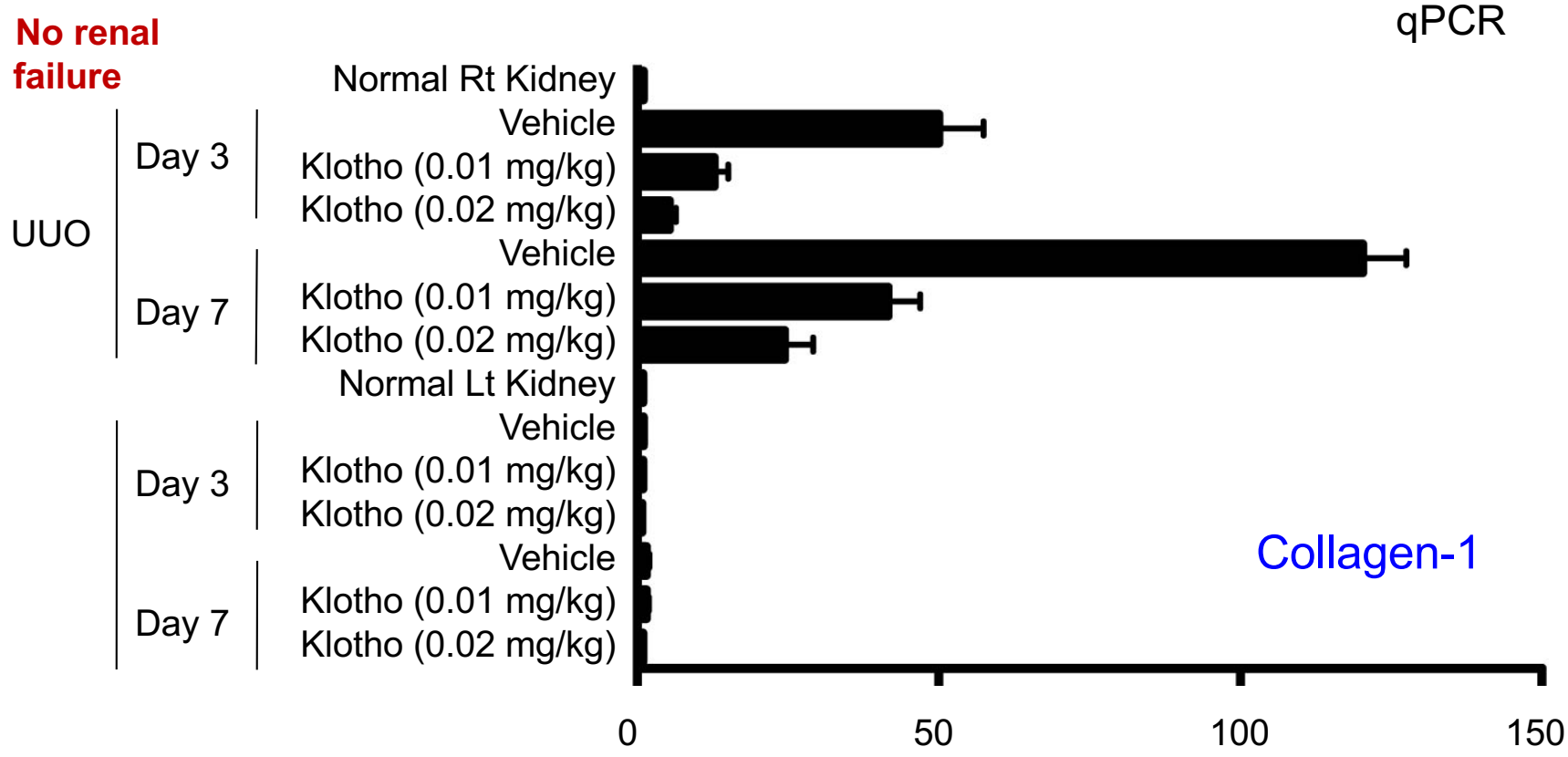


Klotho suppresses fibrosis in an animal model of chronic kidney disease, unilateral ureter obstruction (UUO)

Fibrosis within 7 days

No renal failure



Slide from Dr. Makoto Kuro-o

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Irving J. Bigio

Professor
Biomedical Engineering and Electrical & Computer Engineering, ENG;
Physics, CAS; and Medicine, MED

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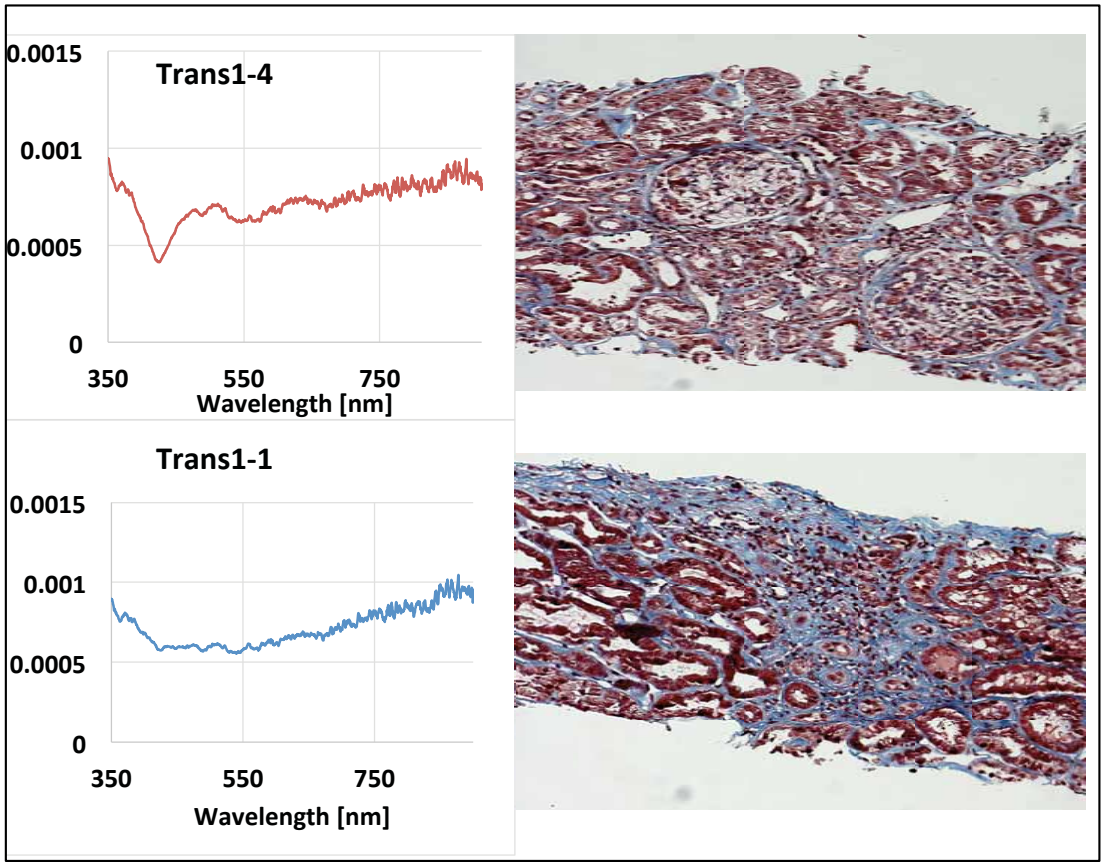
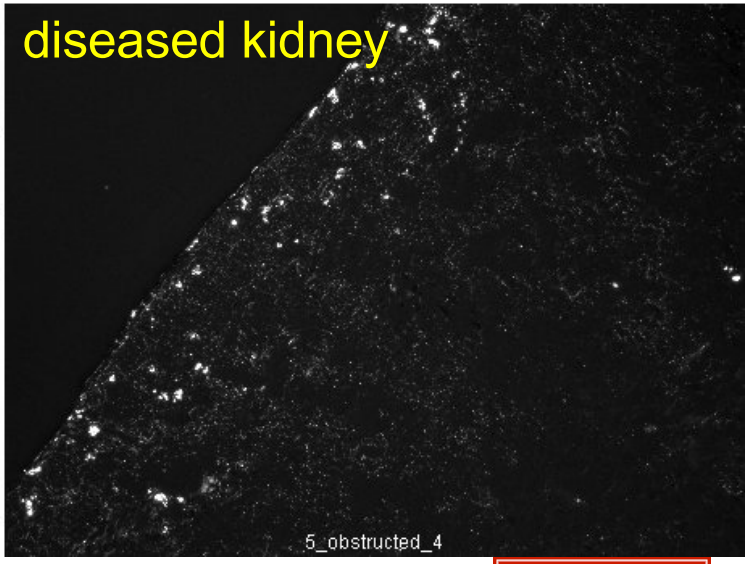
Quantitative optical assessment of fibrosis:

1) Elastic-scattering spectroscopy (ESS): collagen in ECM (fibrosis) exhibits enhanced optical scattering, with short-wavelength bias.



2) Birefringence imaging: collagen is highly birefringent, can be quantitatively imaged with polarized microscopy.



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Activation States of Perivascular Adventitial Fibroblasts

Jeff Browning

Research Professor
Microbiology and Rheumatology, MED

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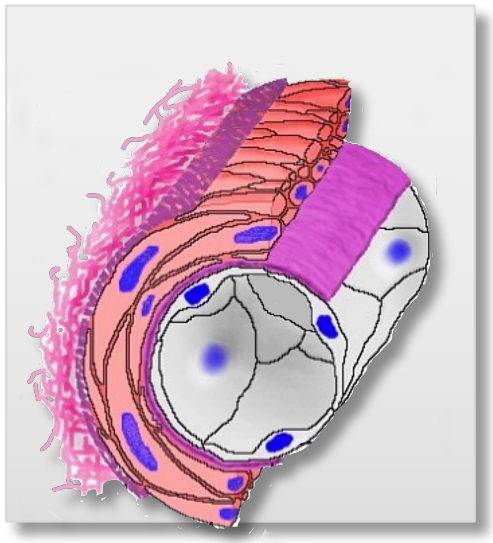
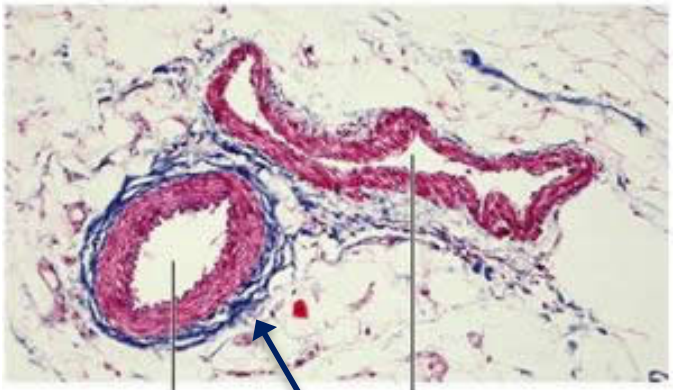
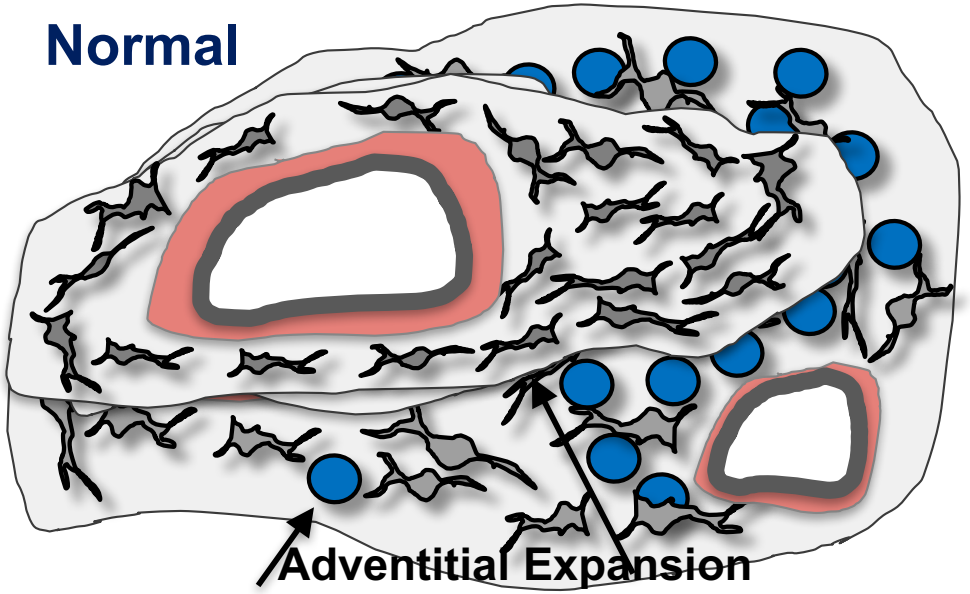
Blood Vessels





Cutaneous Lupus Scleroderma

Normal



Adventitia

Lymphocytic Infiltrate

Collagen Matrix-Rich Niche

Mesenchymal Stem Cells

Monocytes (Resident?)

Adventitial Fibroblasts

Adventitial Fibroblasts gain VCAM expression

Adventitial fibroblasts are beginning to resemble the fibroblastic reticular networks that scaffold the lymphoid organs

Well-poised to orchestrate repair/remodeling

Does the reticular network provide retention signals for lymphocytes?

https://embryology.med.unsw.edu.au/embryology/index.php/HM_Practical_-_Blood_Vessel_Histology

Jacob Joseph

Adjunct Associate Professor
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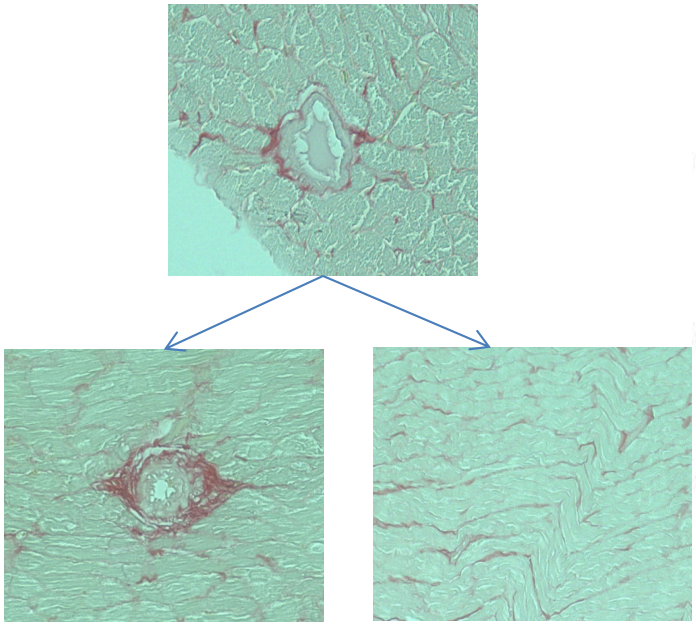
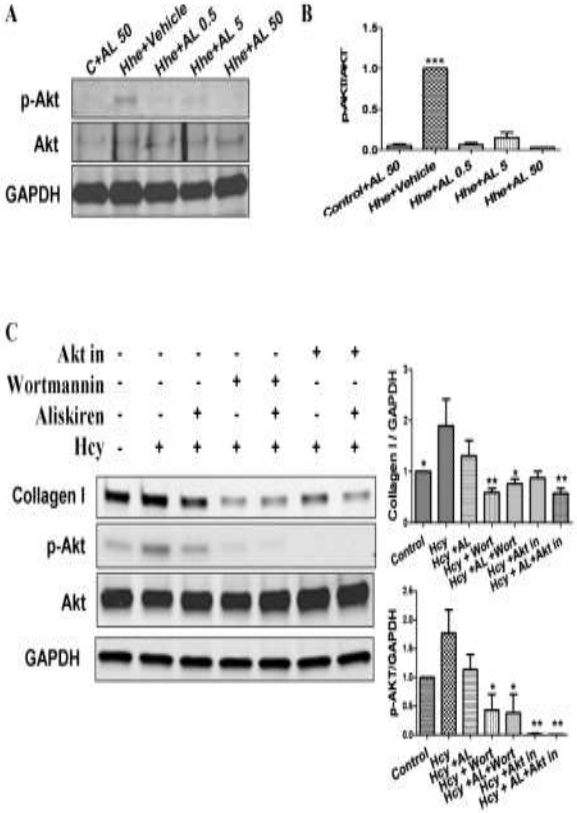
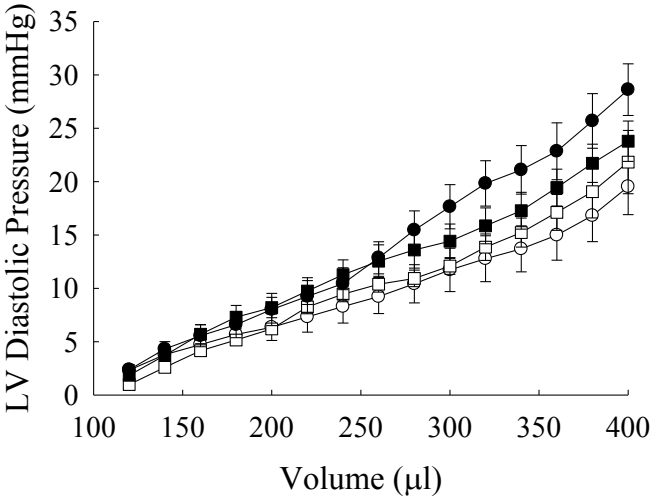
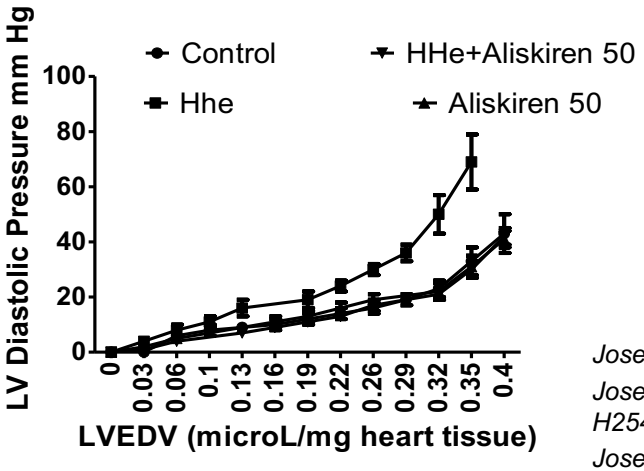
Research on Tap: Connecting Tissues and Investigators

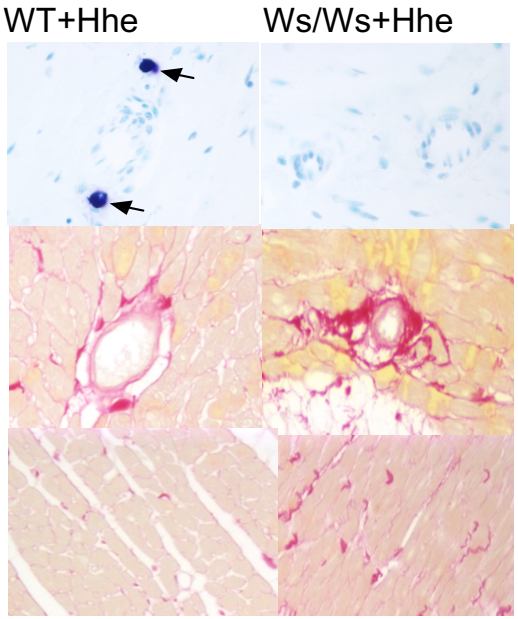


Reactive myocardial fibrosis







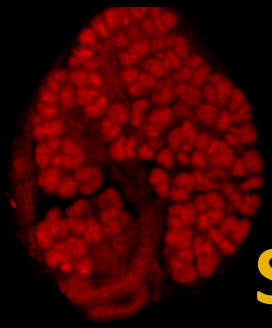


Joseph et al. Am J Physiol 2002; 283: H2567-H2574.

Joseph et al. Am J Physiol Heart Circ Physiol. 2005; 288(5): H2541-5.

Joseph et al. J Heart Lung Transplant. 2008;27(11):1237-41

Zhi et al. PLoS One. 2013 Dec 11;8(12):e81612



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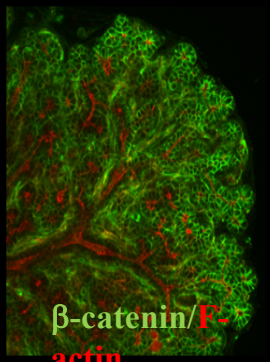
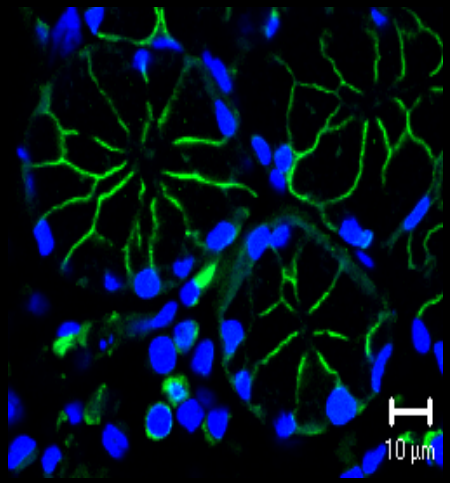
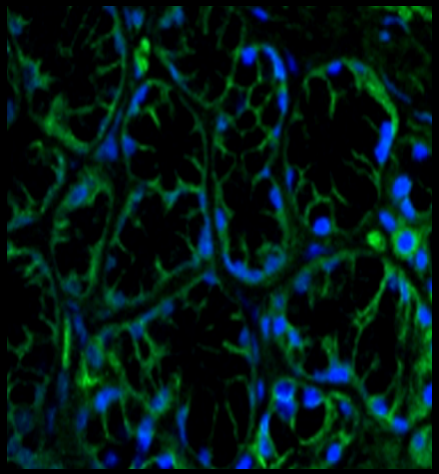


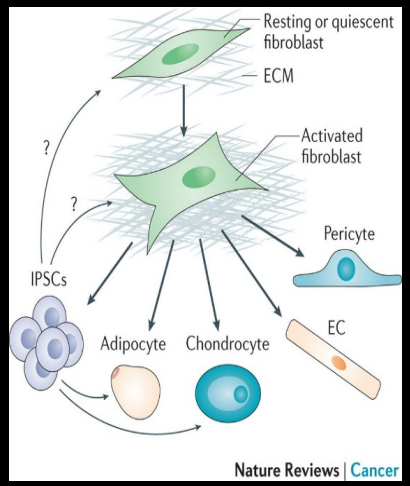
Salivary Gland Repair, Regeneration and Fibrosis

a model for patterned cell and matrix dynamics in branching morphogenesis



Maria A. Kukuruzinska, PhD
Molecular & Cell Biology, GSDM

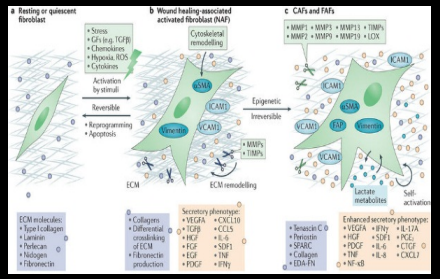




NIDCR National Institute of Dental and Craniofacial Research
National Institutes of Health

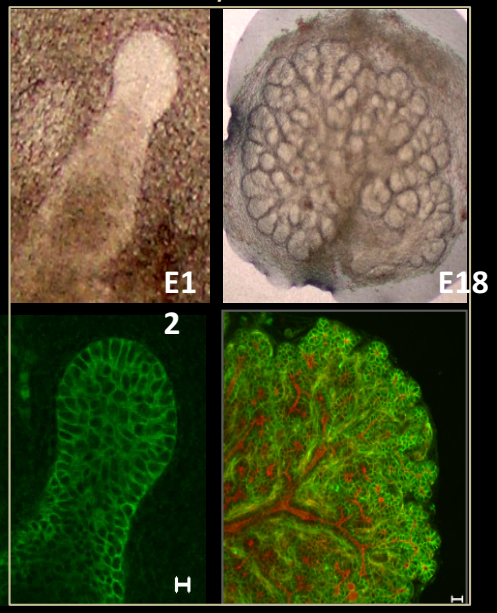
Fibrosis : a multifactorial dynamic disease

Fibroblast activation, Epigentic changes, Atrophy, Metabolic alterations, EMT, Autophagy, ECM remodeling, Innate and adaptive immune responses

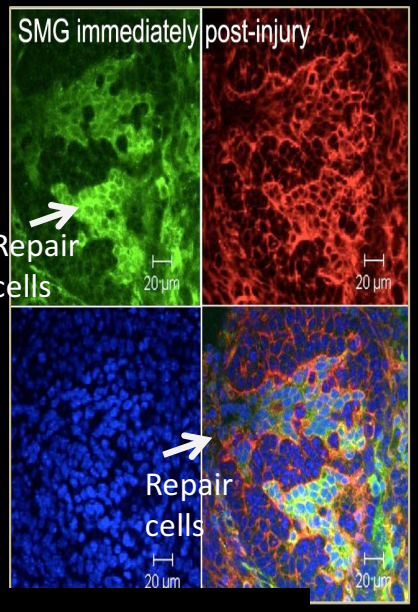


Salivary gland:

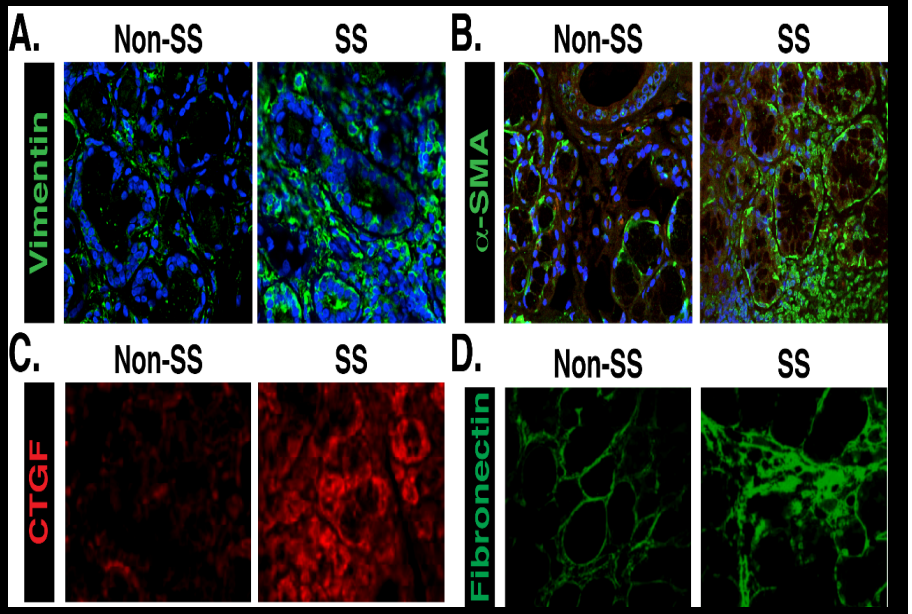
Development

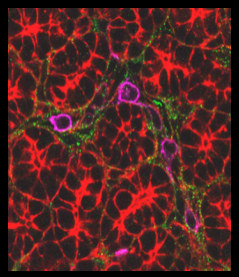


Repair / Regeneration

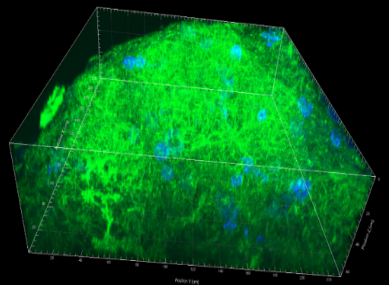


Sjogren's (SS)

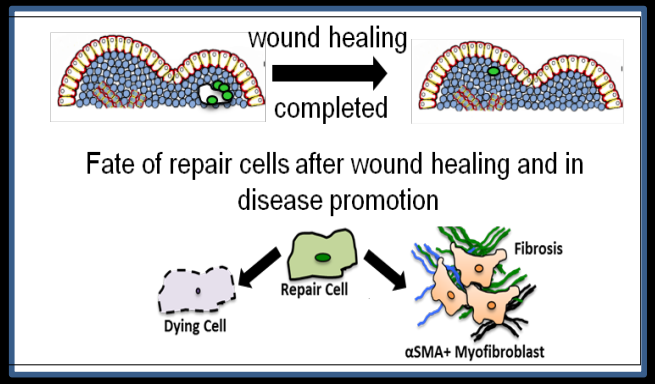




CD68, F-actin



CD45, F-actin



Immune surveillance in the developing salivary gland *J Cell Science, 2013;*

Lab Invest, 2013; Plos Comp Biol, 2016; Sci Reports, 2017 (in revision)

NIDCR National Institute of Dental and Craniofacial Research National Institutes of Health

Regulation of Fibrosis Through Soluble & Mechanical Signals

Matthew Layne

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BOSTON UNIVERSITY

Fibrosis Is Defective ECM Homeostasis

Tissue Injury → Inflammation → Fibroproliferation → Normal Wound Healing

Aortic carboxypeptidase-like protein (ACLP)

Get ACLP "JUST RIGHT"

- ACLP pathways in adipose tissue fibrosis
- Stromal reaction in cancer
- Differentiation of vascular progenitors
- Heritable connective tissue diseases

Fibroblasts *et al* → **Myofibroblasts** → **Signaling**

TGFβ signaling → ECM Mechanotransduction

TGFβ

Mechanical

TOO LITTLE:

Humans: Ehlers Danlos Syndrom

- hypertrophic scars
- wound healing
- vascular disruption

Mice: Delayed wound healing

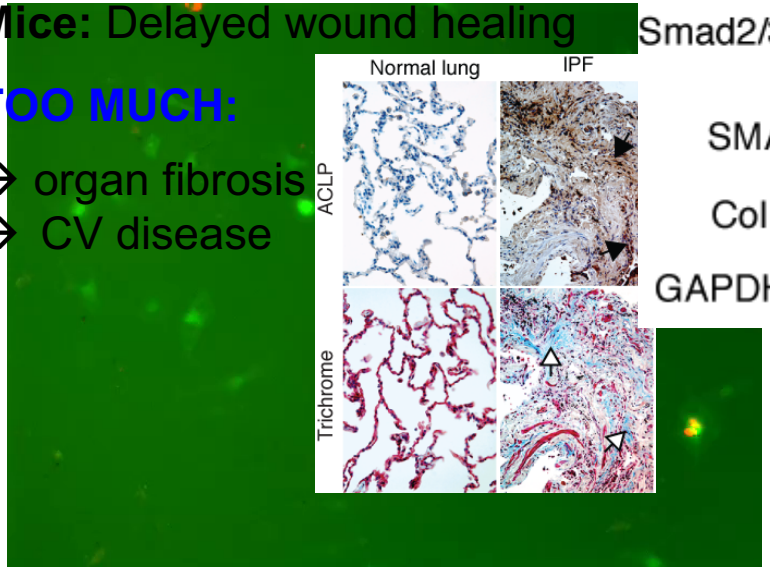
TGFβ + Cell culture plastic (

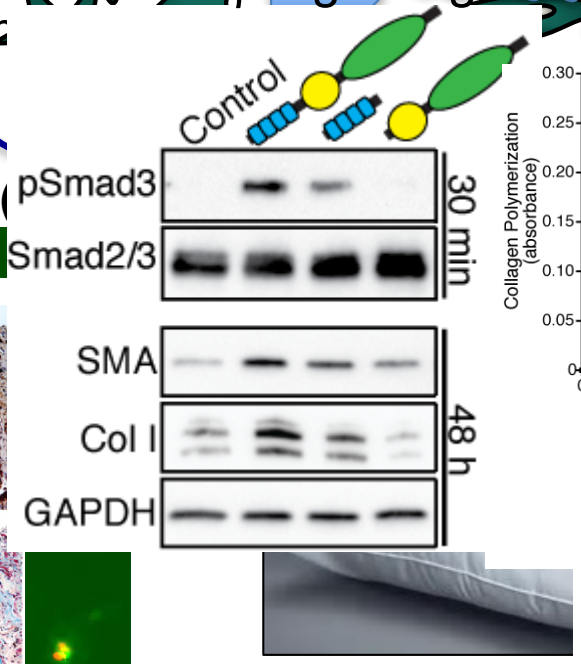
TOO MUCH:

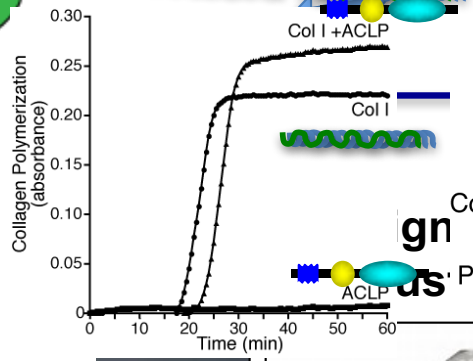
→ organ fibrosis

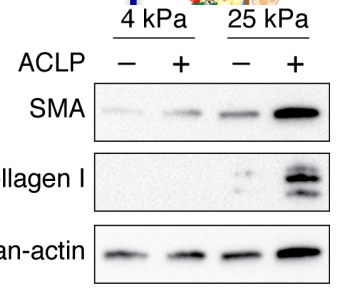
→ CV disease

Collagen SMA

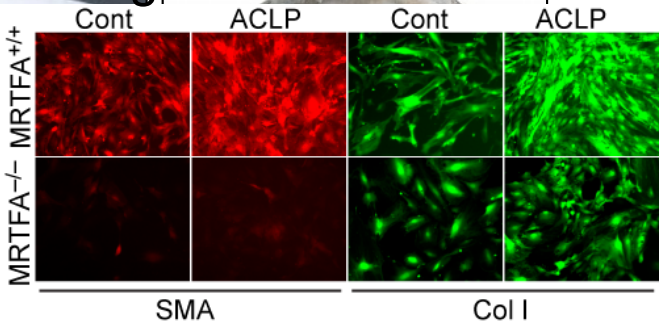








Progenitor differentiation



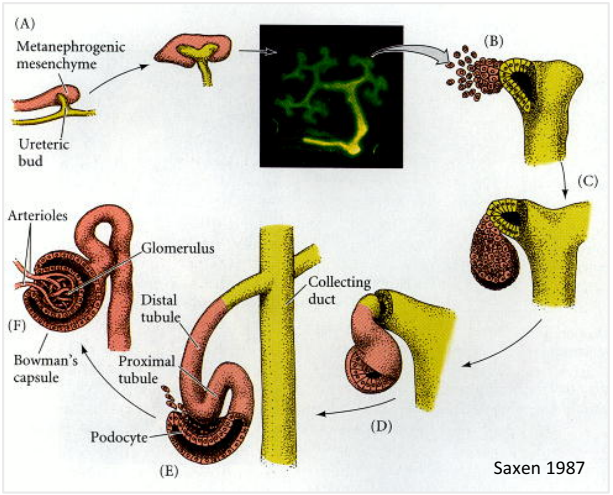
Translational Research to Inform Tissue Fibrosis Mechanism and Drug Discovery

Weining Lu

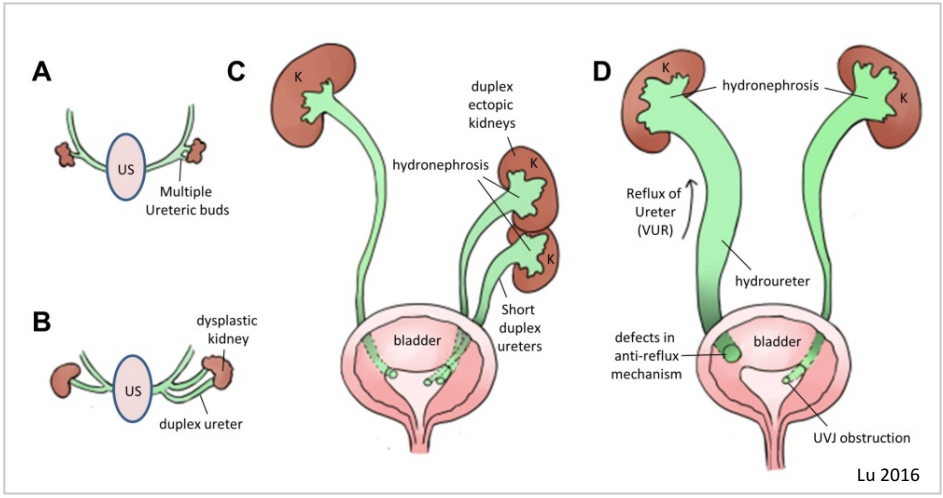
Associate Professor
Medicine and Pathology & Laboratory Medicine, MED

BOSTON UNIVERSITY

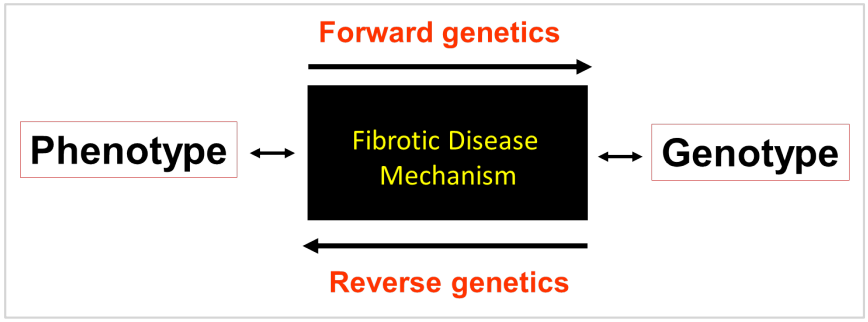
LU LAB: Apply translational developmental genetics to study fibrotic disease mechanism



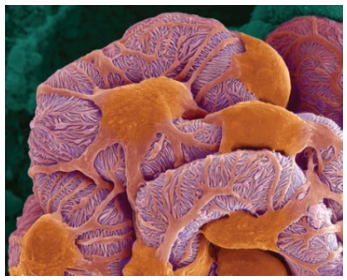
Normal kidney development



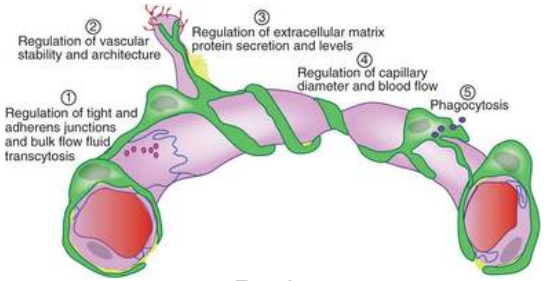
Congenital anomalies of the kidney and urinary tract (CAKUT)



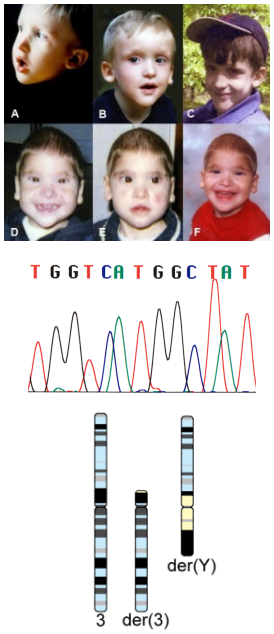
Molecular genetics approach to study fibrotic disease mechanism



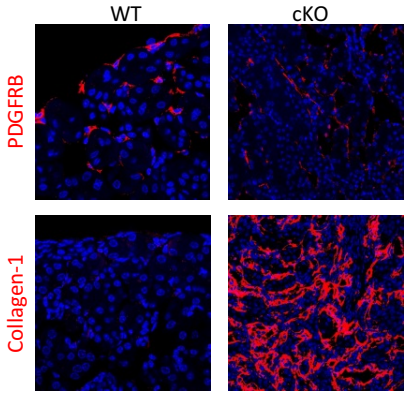
Podocyte



Pericyte



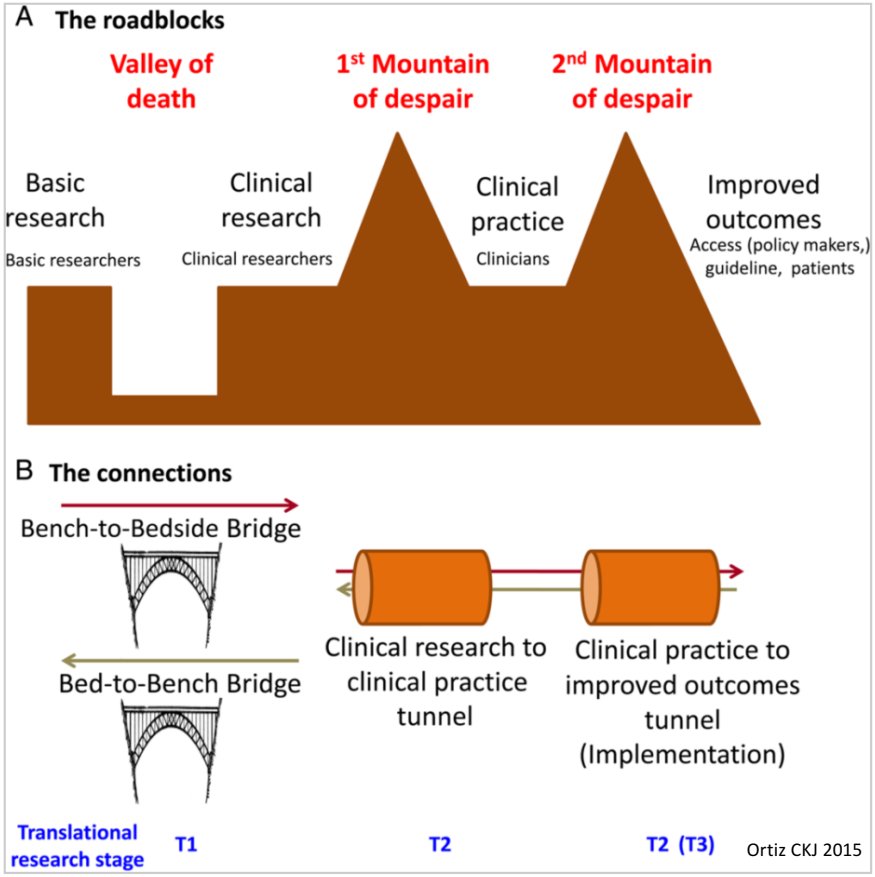
Human genetics



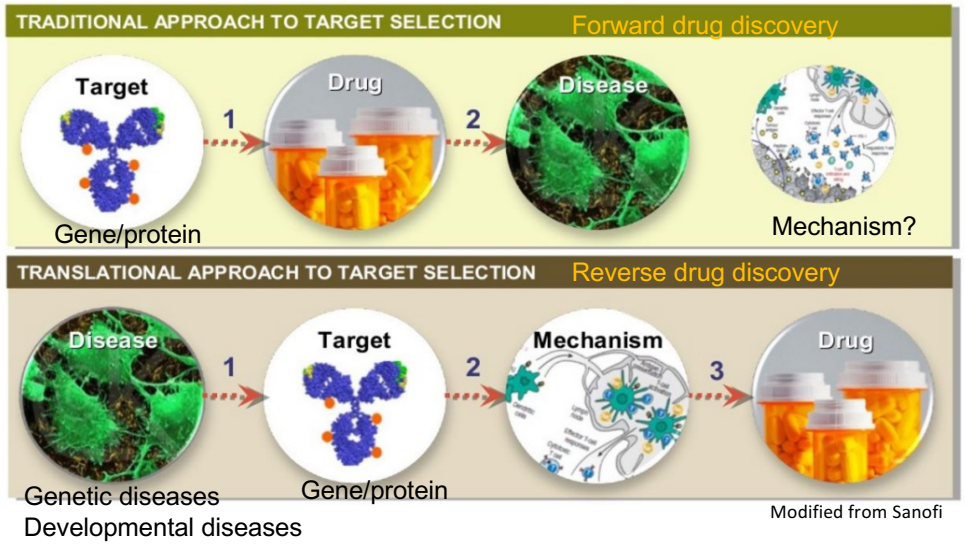
Mouse genetics

ZEB and SLIT-ROBO signaling pathways

LU LAB: Translational research approach to novel drug discovery in fibrotic disease



Translational research



Translational approach to drug discovery

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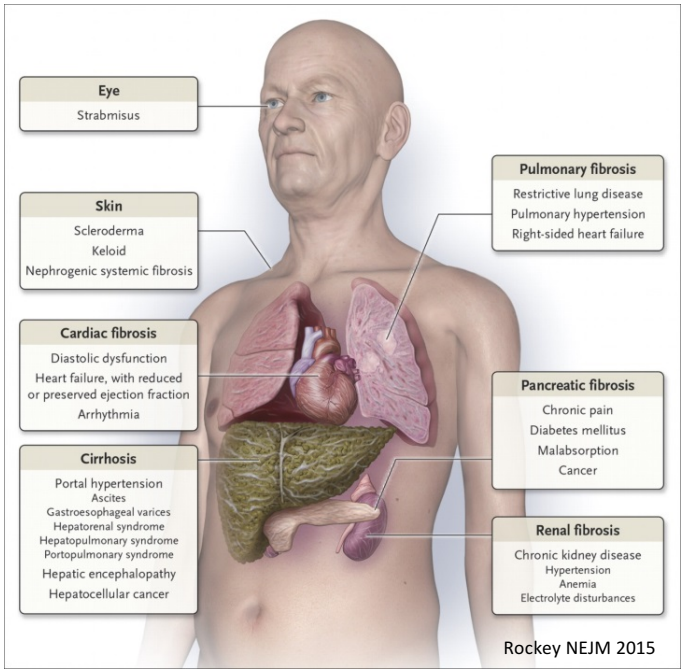
EXCEPTIONAL CARE. WITHOUT EXCEPTION.

Pfizer CTI
Centers for Therapeutic Innovation
A Pfizer-supported collaboration

Academic-industry collaboration accelerates novel drug discovery



Novel drugs for fibrotic disease

The Impact of a Bone Marrow Fibrotic Niche on Blood Cell Development

Katya Ravid

Professor
Medicine and Biochemistry, MED
Whitaker Cardiovascular Institute

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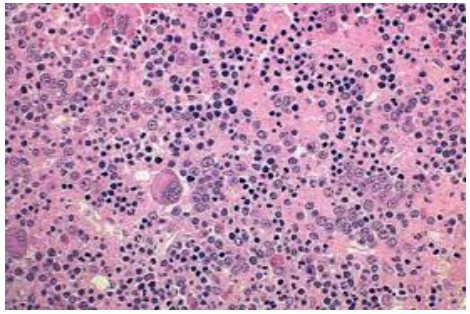
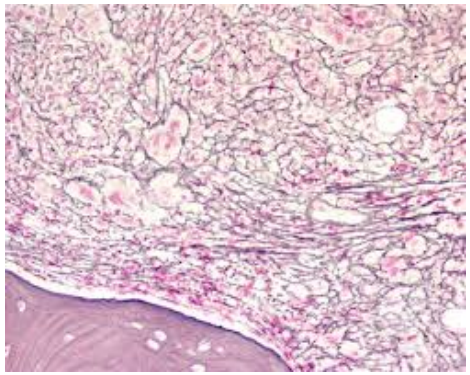
K.Ravid (12/6/17)

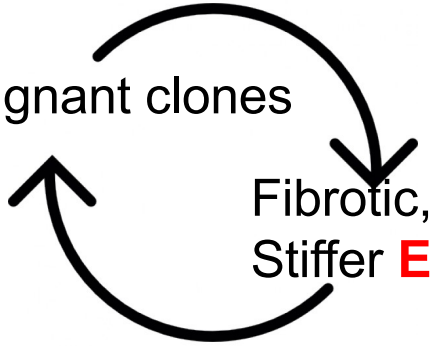
The impact of a bone marrow fibrotic niche on blood cell development

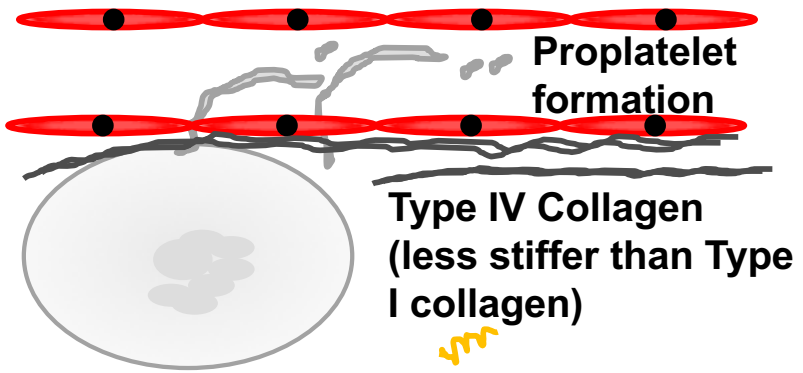
Normal bone marrow (BM)



Fibrotic BM







- Reticulin fibers made of secreted type III **collagen**
- Secreted **Lysyl Oxidase** is associated with a cross-linked **stiffer collagen**

Signaling from the ECM to the cell

ECM->**?** ->FAK->RhoA->ROCK->YAP/TAZ->gene expression

We identified the following sensors of a stiffer ECM towards regulation of platelet production:

- Transient receptor potential cation channel subfamily V member 4 (**TRPV4**)
- **Piezo 1/2** mechanosensetive receptors
- *Inquiry: How do these mechanosensors control BM cells and platelet development?*

ECM->***Mechanosensitive ion channels*** **->, <-** ? YAP/TAZ

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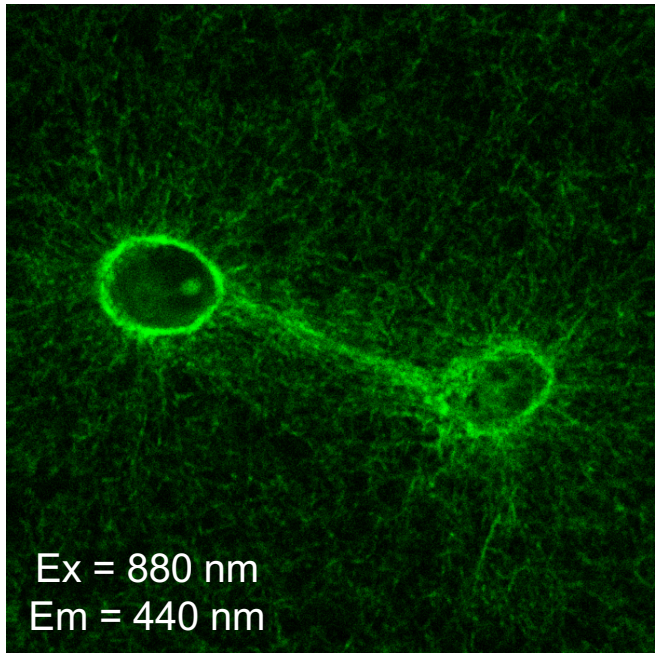
Darren Roblyer

Assistant Professor
Biomedical Engineering, MED

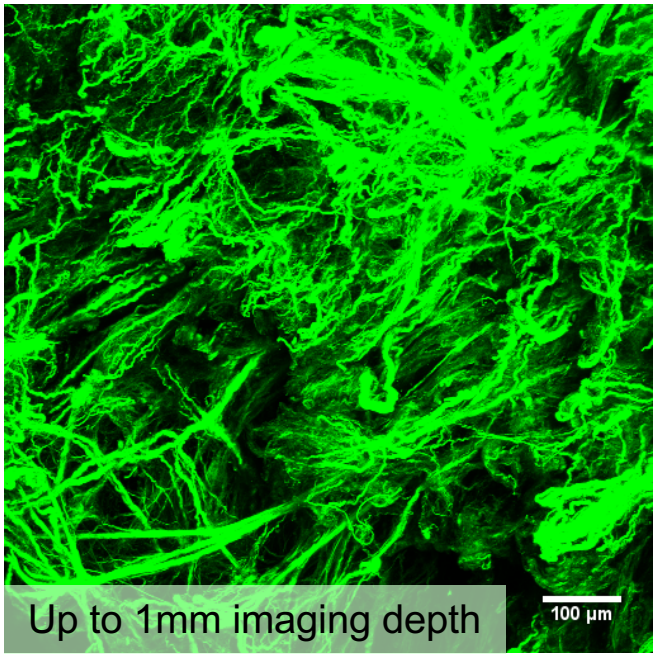
Multiphoton label-free imaging of fibrosis

Second Harmonic Generation Imaging

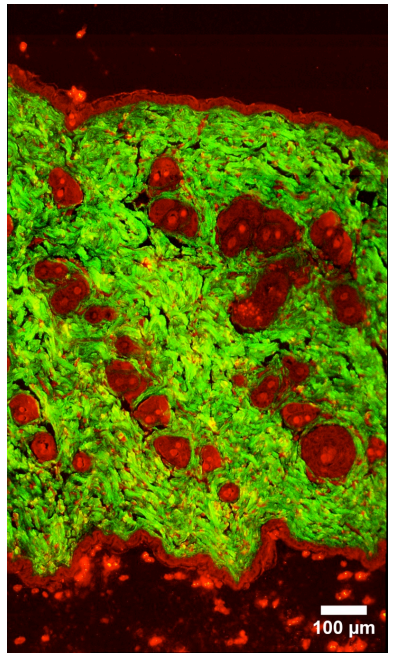
MDA-MB-231 (tumor) cells



Intravital tumor collagen



Mouse skin slide



Angiotensin Fibrosis Model
Sample from Trojanowska Lab

Label free imaging of type I and II collagen
fixed or fresh samples
In vivo (skin, window chamber)
layer thicknesses, orientation, 3-D volumes
New! <5 pubs on fibrosis (mostly liver fibrosis)

Green = collagen
Red = FAD

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An Agent-Based Network Model of Pulmonary Fibrosis Development

Béla Suki

Professor

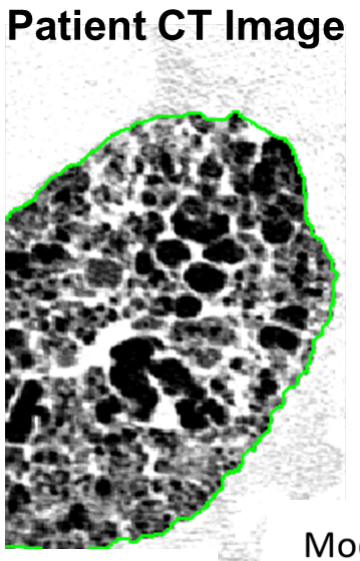
Tyler J. Wellman

Biomedical Engineering, ENG

Research on Tap: Connecting Tissues and Investigators

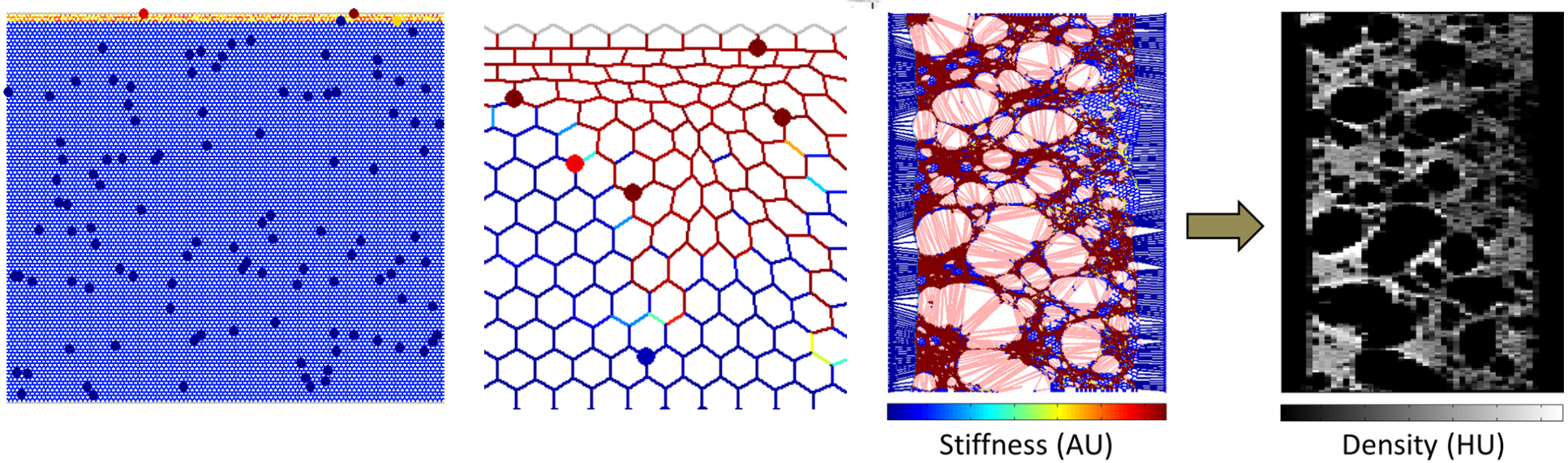
Motivation

- Idiopathic Pulmonary Fibrosis (IPF) lungs exhibit distinct structural changes as a result of pathologic changes in cell behavior:
  - Subpleural Honeycombing / Cyst formation
  - Traction Bronchiectasis
  - Reduction in Lung Volume, Compliance



Rationale

- The known cell behaviors should lead to the observed changes in lung structure and associated mechanical dysfunction.
  - Resistance to Apoptosis
  - Collagen Expression / Deposition
  - Cell Motility / Invasiveness



Conclusion: 1) Cell activation and migration on stiff tissue and 2) Mechanical failure can explain deterioration in lung structure and function in pulmonary fibrosis.

Boston University Office of the Vice President and Associate Provost for Research

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Multifunctional Lysyl Oxidases and Fibrosis

Philip Trackman

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Molecular & Cell Biology, GSDM

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Lysyl Oxidases

- Gene family made up of five genes: *LOX, LOXL1 – LOXL4*
- Critically required for ECM biosynthesis:
  Extracellular maturation of collagens & elastin via oxidation of lysine residues.
- The Three Bears and Goldi-*LOX*:
  Too little: poor connective tissue structure: osteolathyrism, aneurisms
  Too much: excess collagen accumulation and fibrosis, metastasis
- Novel substrates and functions relevant to fibrosis:
  PDGFR-β (increased cell proliferation, chemotaxis?)
  αV-integrin (increased fibroblast adhesion, unpublished)

- Propeptide regions direct proenzymes to ECM molecules for activation by procollagen C-proteinases and function: fibulins, fibrillin, tropoelastin, periostin, fibronectin, others? *Implies importance of functional extracellular protein complexes.*
- LOX-PP is released, has independent functions and binding partners: tumor growth inhibitor, has no enzyme activity. *Some functions are independent of enzyme activity.*

- Novel small pharmacologic inhibitors are being developed, and some are available from companies.
- Genetic models: We are creating a Floxed LOX mouse by CRISPR/Cas9 technology for tissue-specific knockout studies.

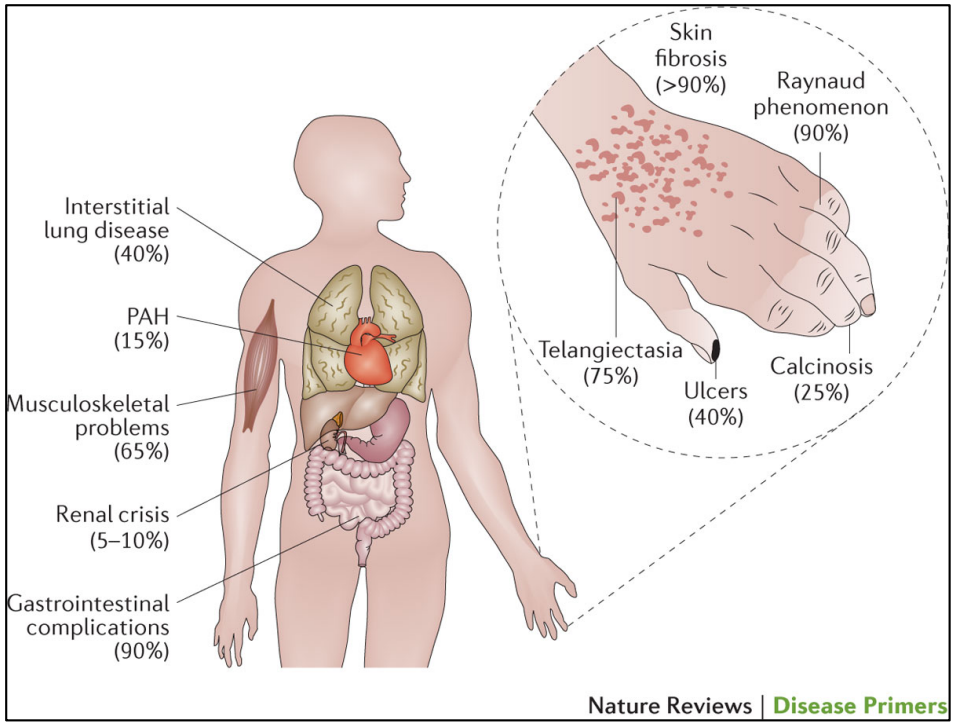
Beyond Fibrosis: The Challenges of Scleroderma

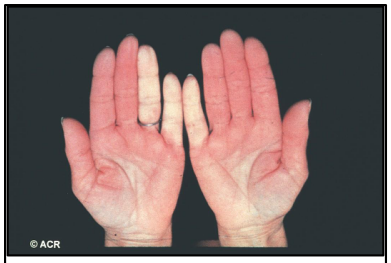
Maria Trojanowska

Professor
Medicine, MED;
Director, Arthritis Center

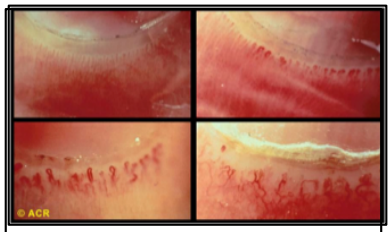
BOSTON UNIVERSITY

Organ complications associated with systemic sclerosis (scleroderma)

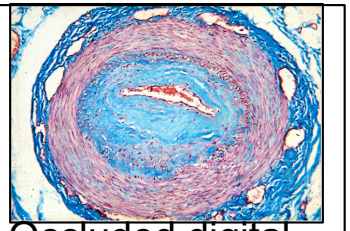


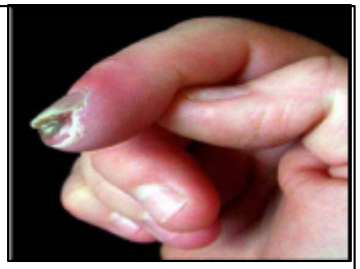


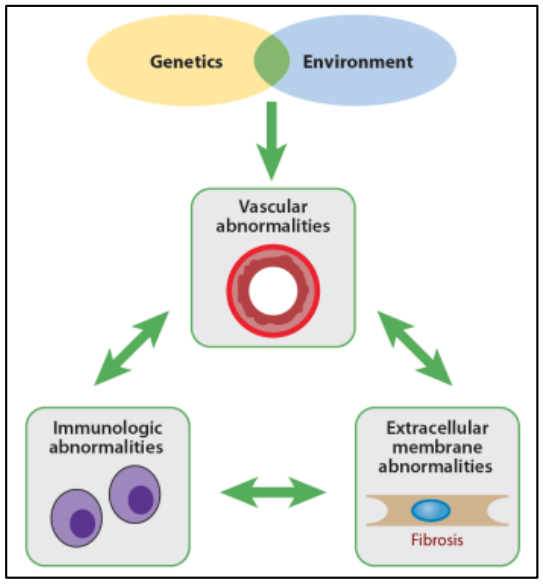
Raynaud's syndrome

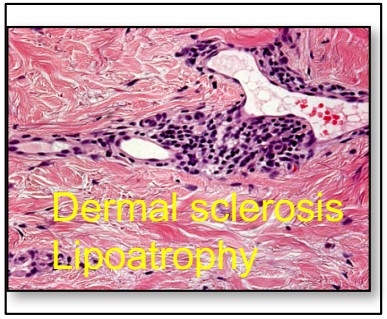


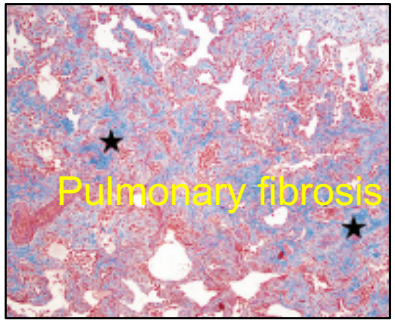
Abnormal capillaries

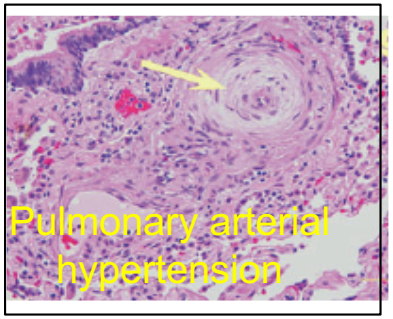

Occluded digital artery


Digital ulcers







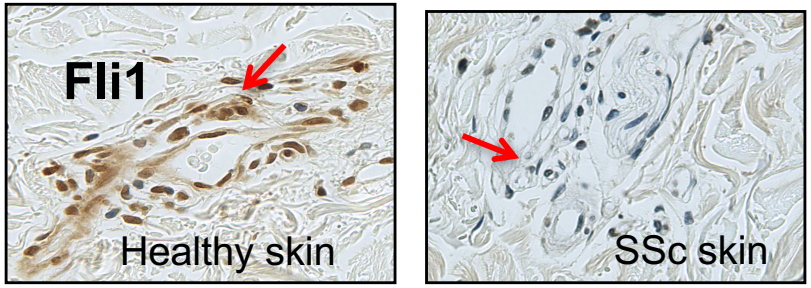


FUTURE CHALLENGES:

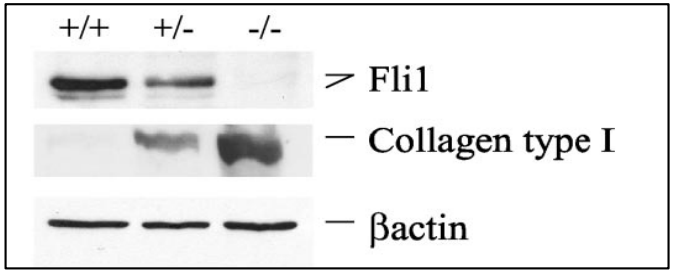
- To better understand the interactions among different disease components
- To find a cure!

Disease models

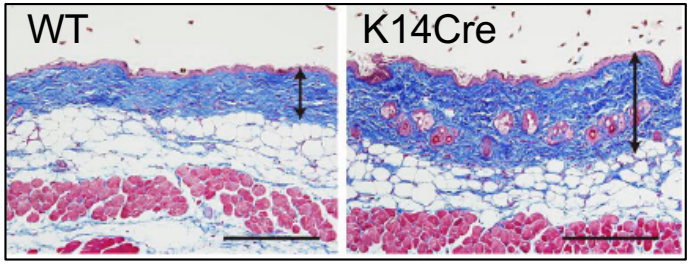
Transcription factor Fli1, member of the Ets family (Friend murine leukemia integration-1)



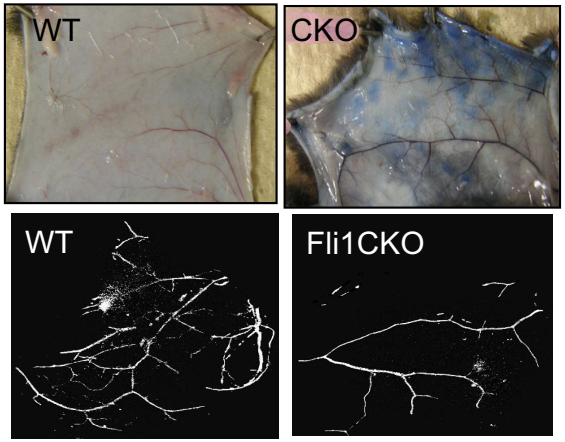
Fli1 plays a central role in ECM regulation



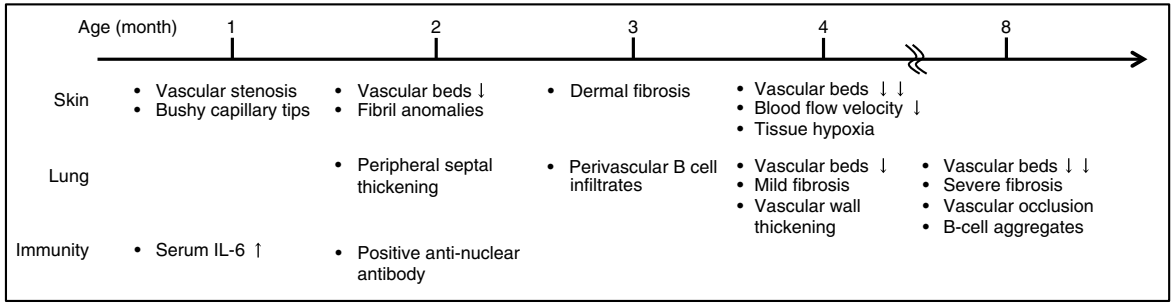
Epithelial Fli1 deficiency drives autoimmunity and fibrosis



Endothelial Fli1 deficiency phenocopy SSc vasculopathy



Simultaneous downregulation of Fli1 and KLF5 reproduces key features of scleroderma



YAP/TAZ Signaling in Fibrosis

Bob Varelas

Associate Professor
Biochemistry, MED

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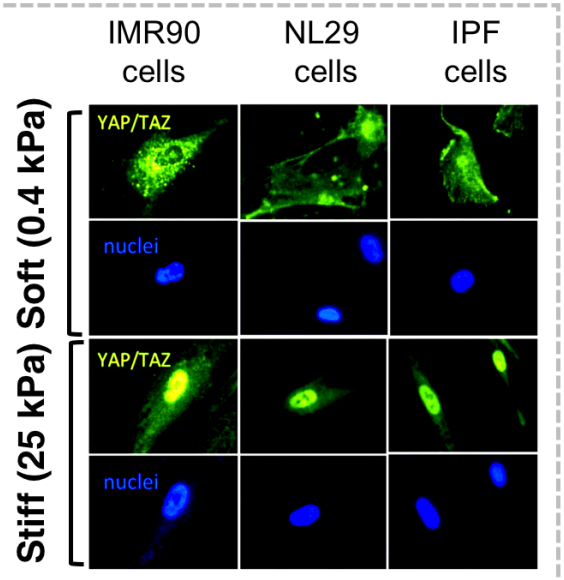
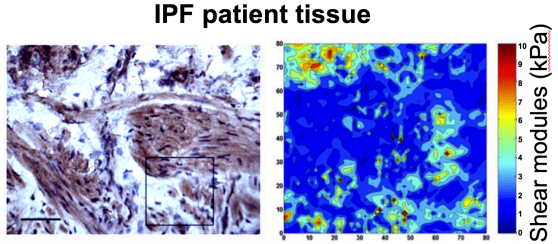
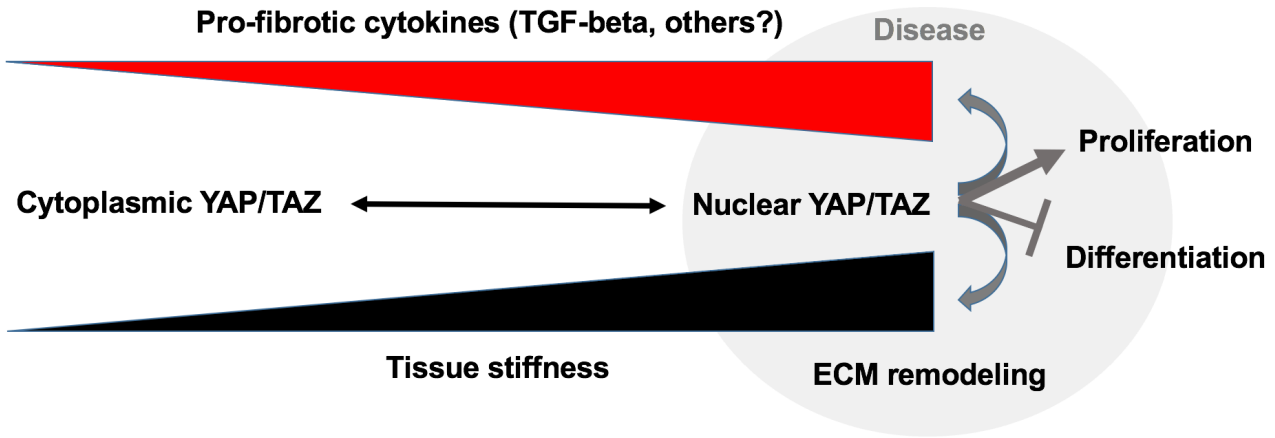
Aberrant YAP/TAZ signaling drives pro-fibrotic disease phenotypes



Liu et al. *J Physiol Lung Cell Mol Physiol* 2015





Ongoing/Future directions:

- Test how altering YAP/TAZ activity affects fibrotic processes in vivo
- Define transcriptional events regulated by YAP/TAZ
- Characterize secreted factors regulated by YAP/TAZ
- Identify downstream YAP/TAZ-regulated effectors important for promoting fibrosis (biomarkers/therapy)

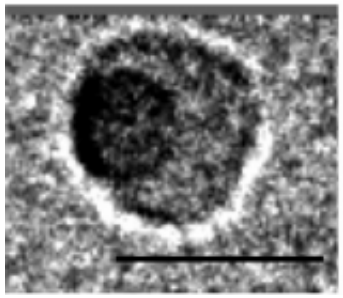
The Role of ECM in Fibrosis: Collaborative Studies

Joyce Y. Wong

Professor
Biomedical Engineering and Materials Science & Engineering, ENG

BOSTON UNIVERSITY

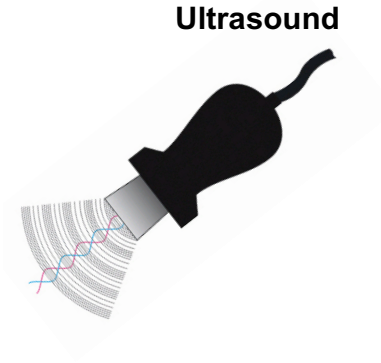
Theranostics ARC (Herrera, Ruiz-Opazo)
(Zhong)



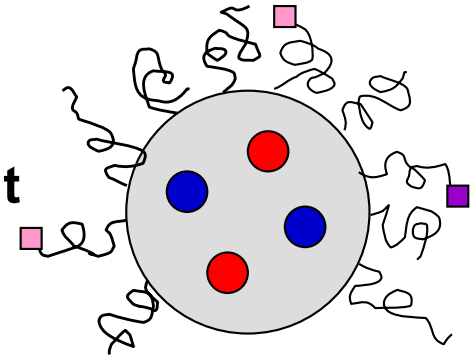
MRI contrast agent

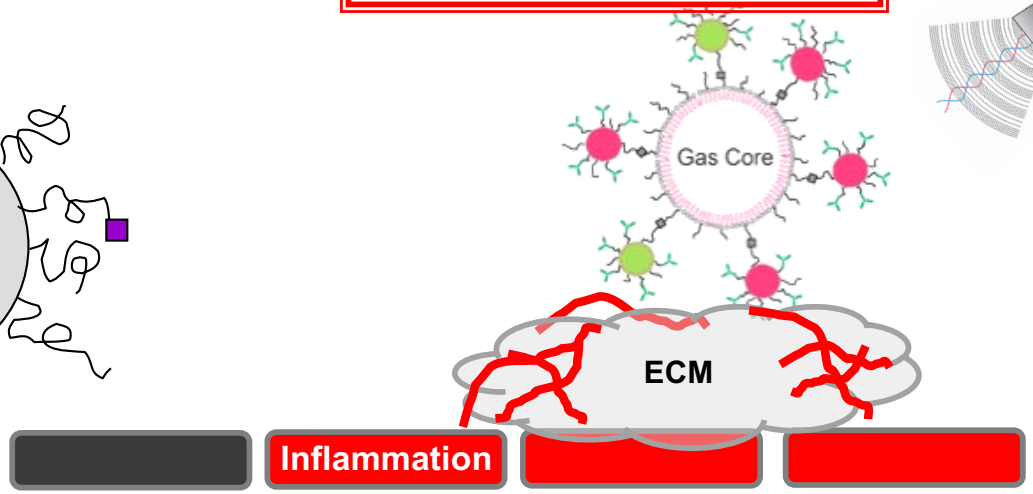
Can we 'see' fibrosis (ECM)?

Detection Modality
- **ultrasound**
- **MRI**
- **CT**
- **fluorescence**



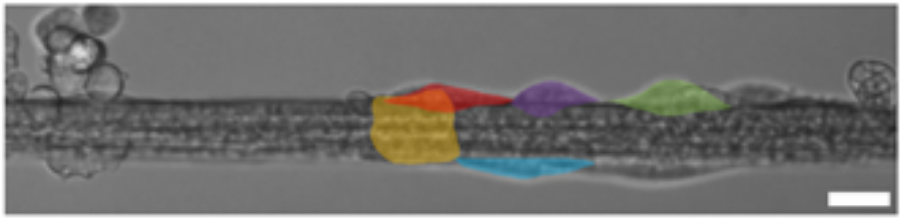
tether
ligand
contrast agent
drug





(Holt, Kasotakis, Nagy)

We can make ECM alloy fibers:



(Smith)

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