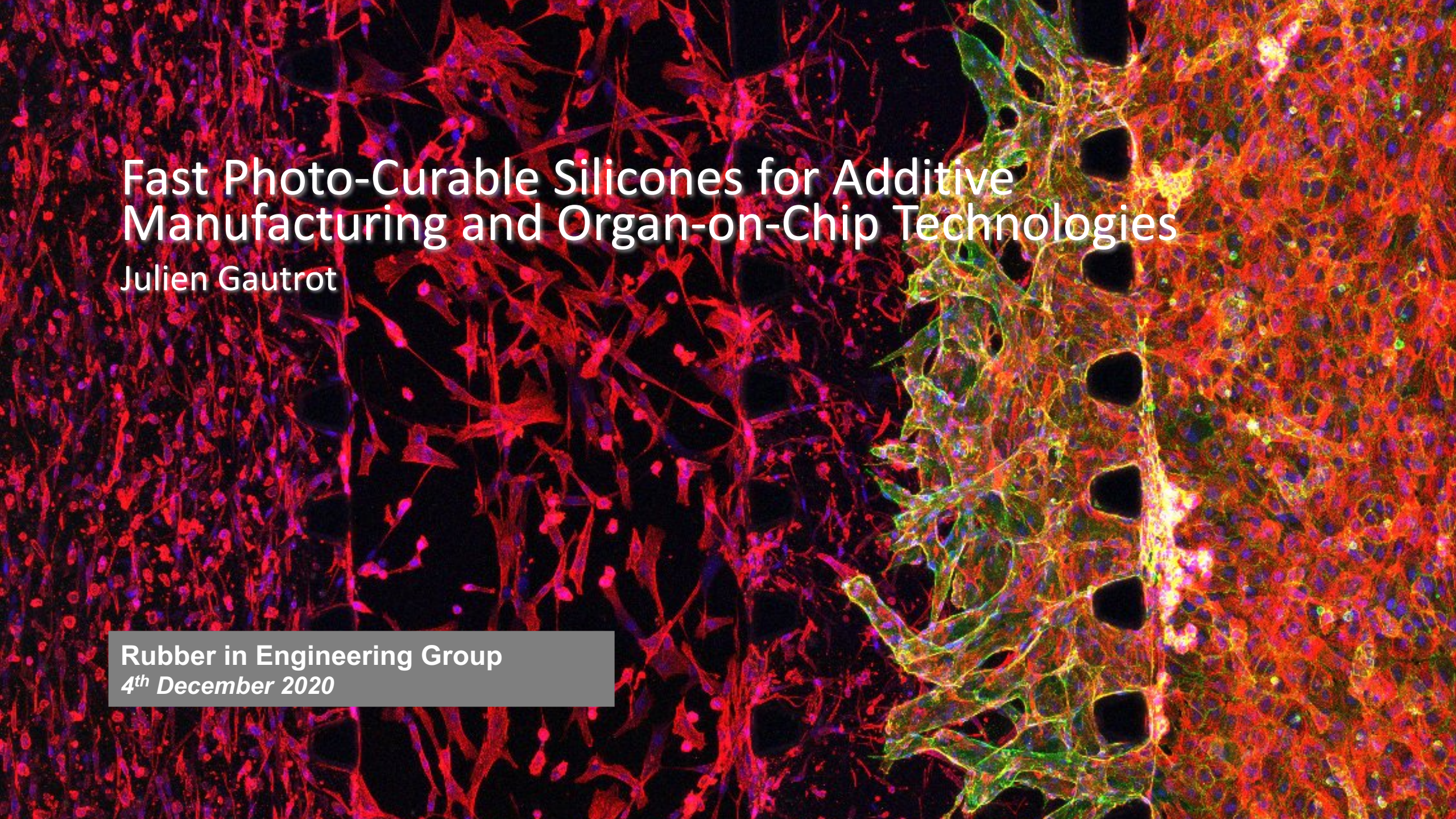
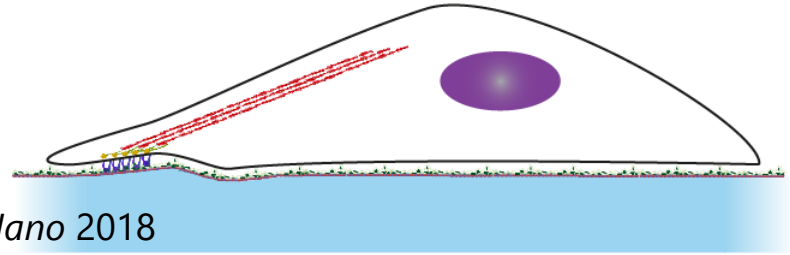


Fast Photo-Curable Silicones for Additive Manufacturing and Organ-on-Chip Technologies

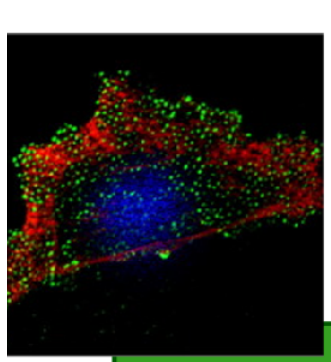
Julien Gautrot

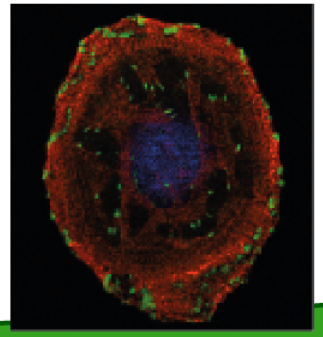
Rubber in Engineering Group
4th December 2020

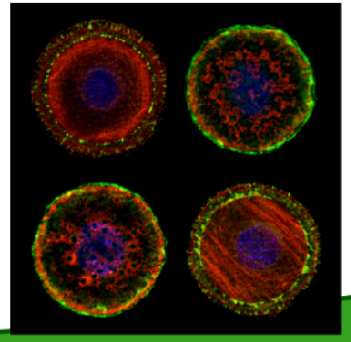
Engineering the Cell Microenvironment at Multiple Scales

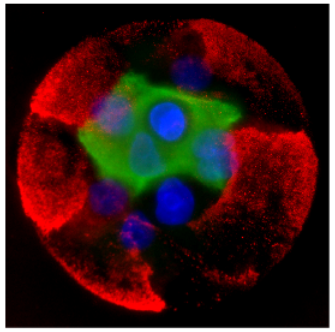


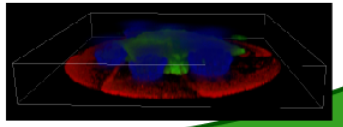
Kong et al. *ACS Nano* 2018
Kong et al. *Nano Letters* 2018
Trappmann, Gautrot et al. *Nat. Mater.* 2012
Costa et al. Acta Biomater. 2014
Gautrot et al. *Nano Letters* 2014

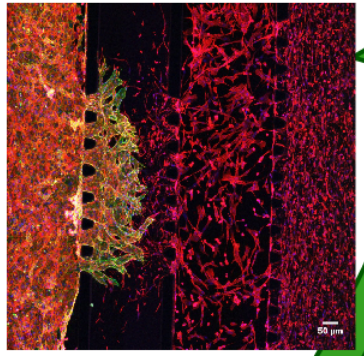


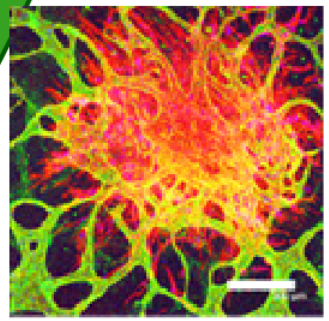


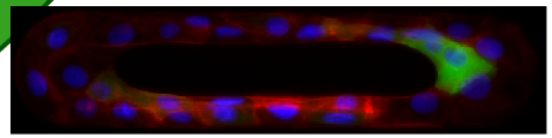


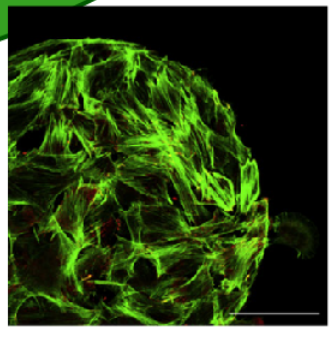


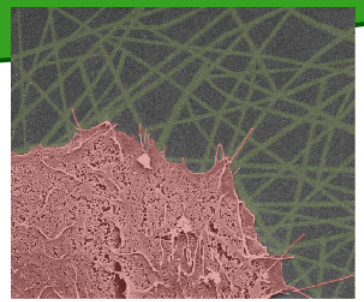


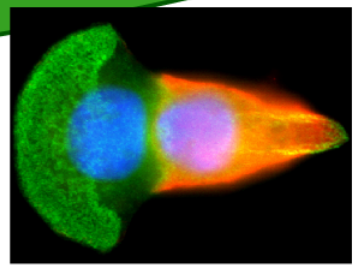










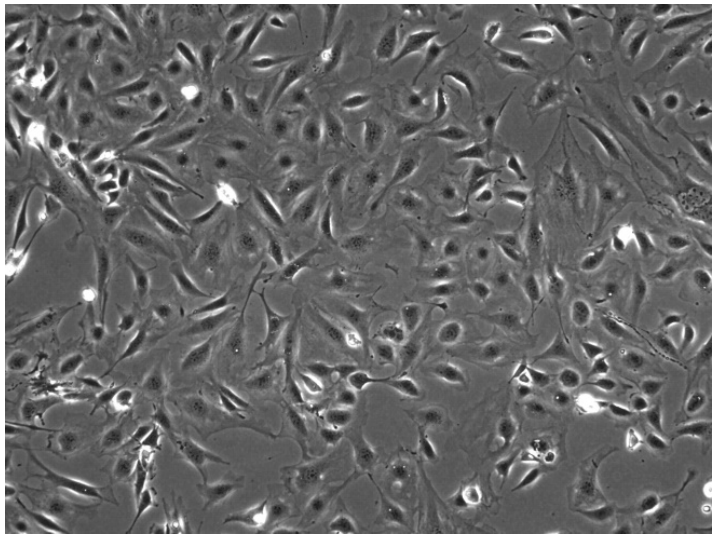


100 nm
1 µm
100 µm
1 mm

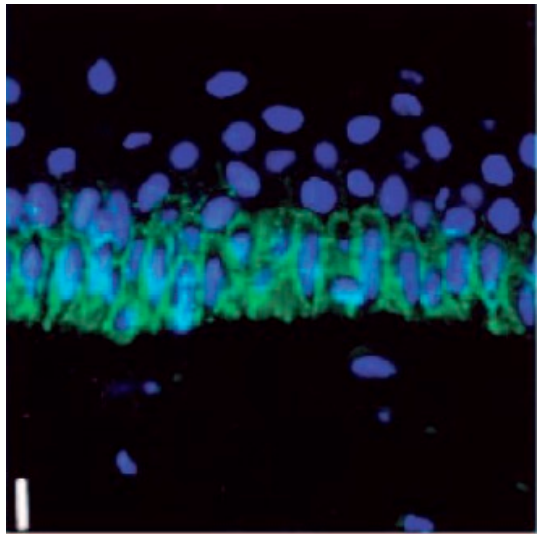
Colak et al. Biomacromolecules 2018
Di Cio et al. *Acta Biomat.* 2016
Di Cio et al. *Acta Biomat.* 2017
Tan et al. Integ. Biol. 2013
Connelly et al. *Nat. Cell Biol.* 2010
Gautrot et al. *Biomaterials* 2012

Cells in Culture Do not Look Like Cells *in Vivo*

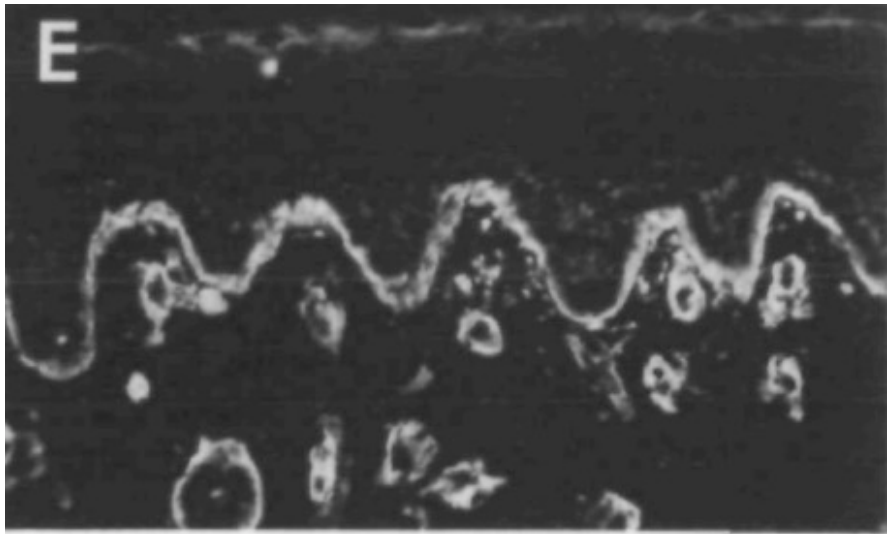
2D In Vitro Culture



In Vivo



Niessen et al. J. Cell Sci (1996), *109*, 1695.

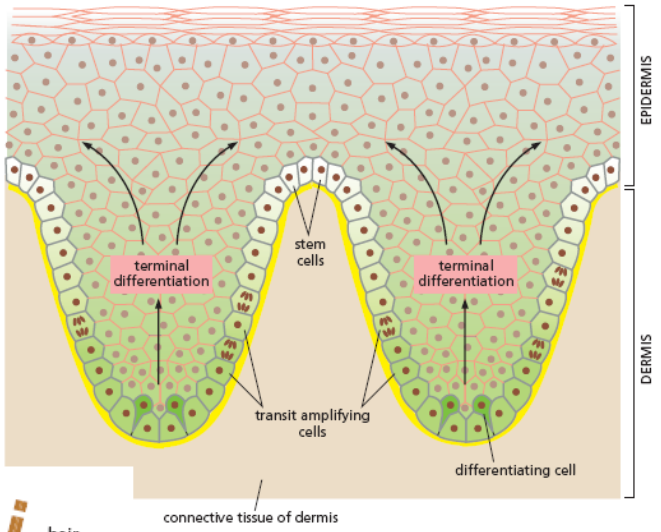


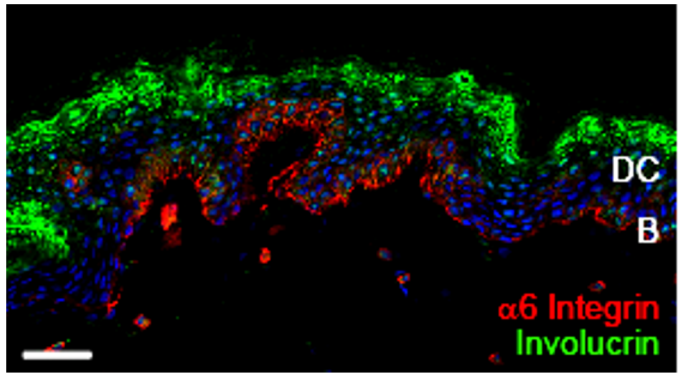
Herle et al. Development (1991), *112*, 193.

- Cells and tissues in vivo have reproducible shapes, size and geometries.
- In 2D cultures, cell shape is unconstrained.

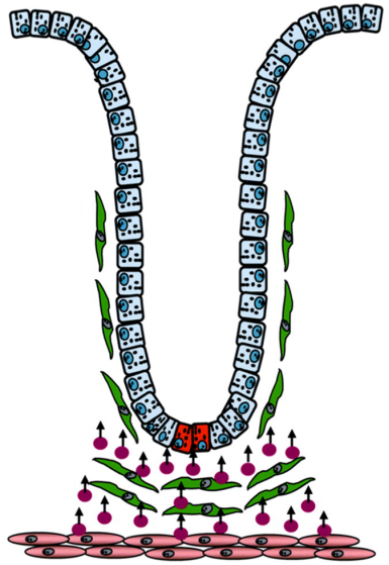
Geometry, Structure and Function

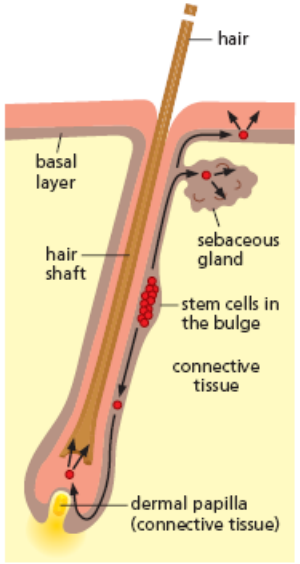
Skin - Epidermis



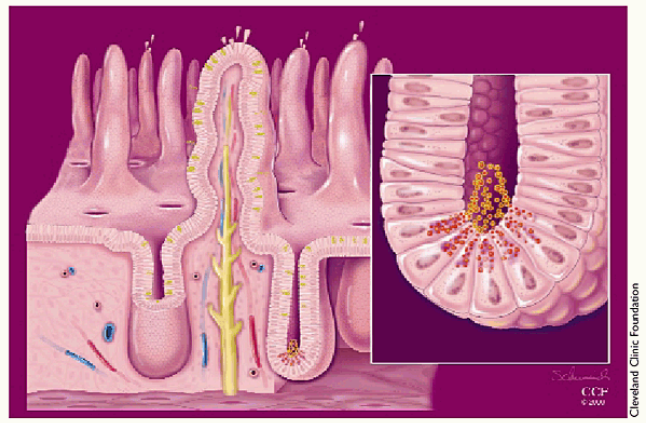


Intestine

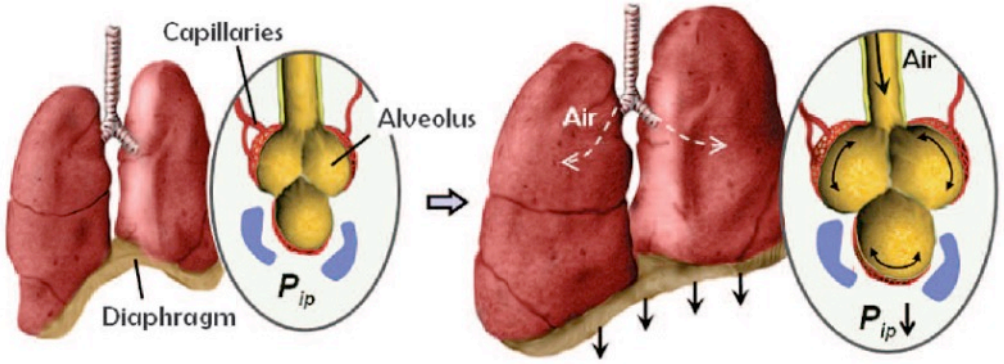


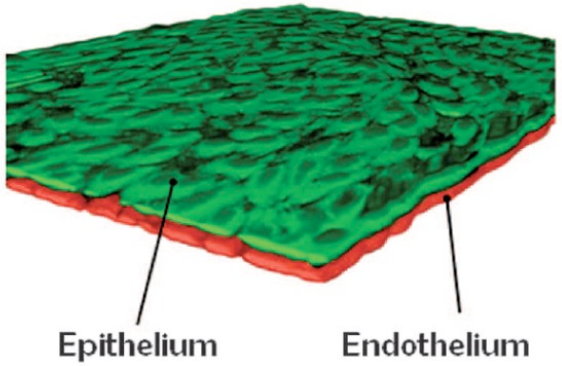


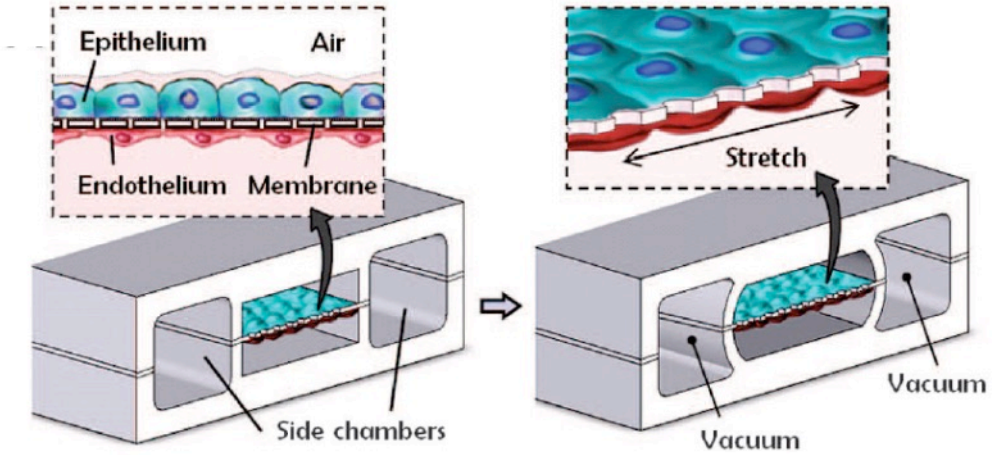
- Stem cells reside in well defined locations.
- The architecture of the tissue and that of the "niche" are important to ensure proper functioning of stem cells and tissue hometostasis.

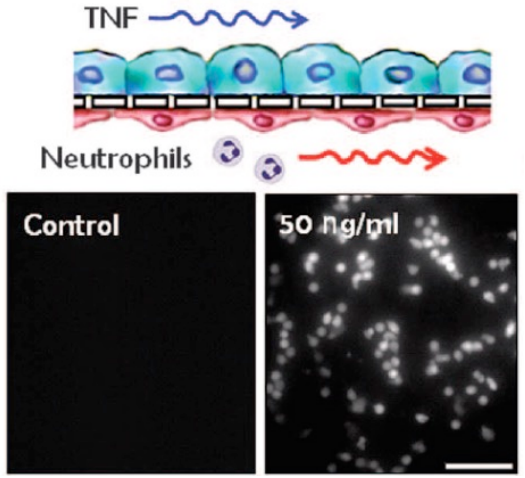


Recreating Higher Level Structure and Function





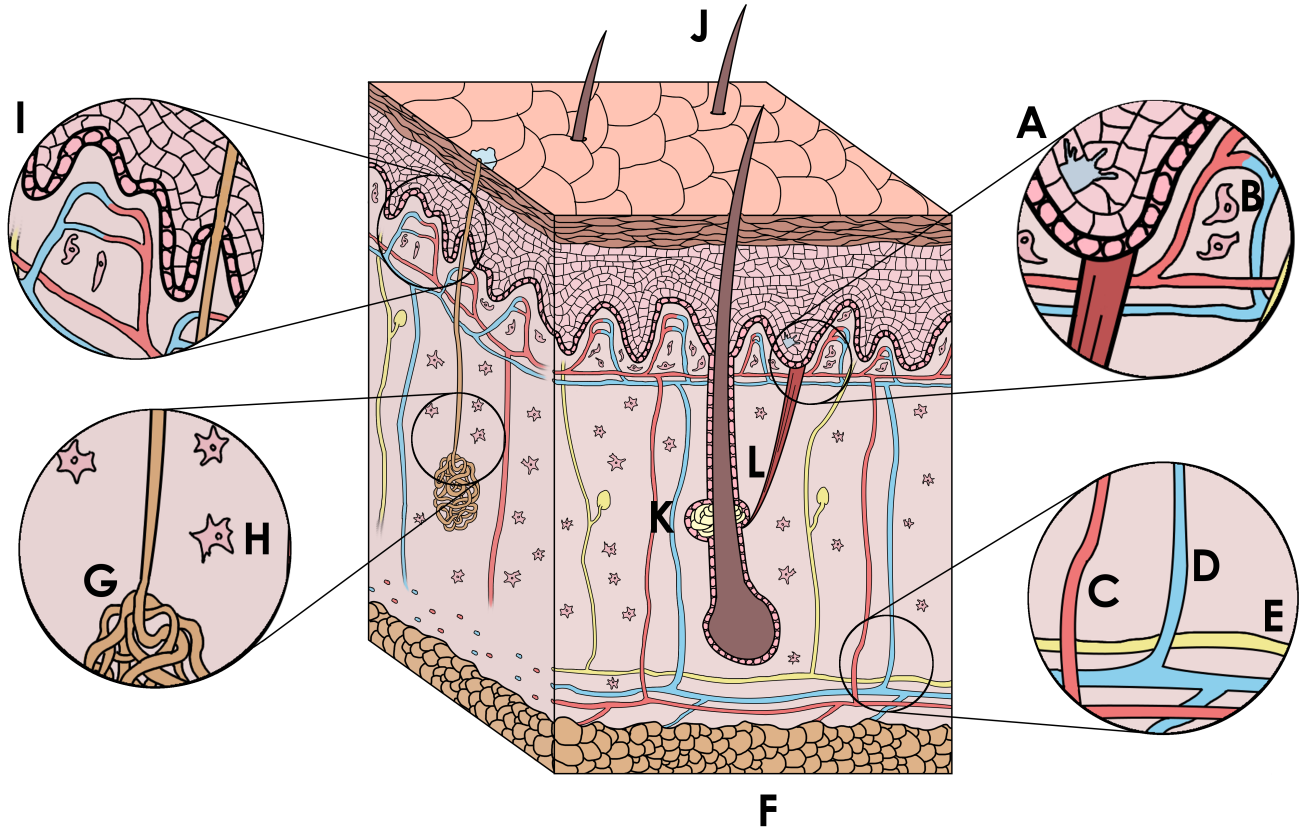




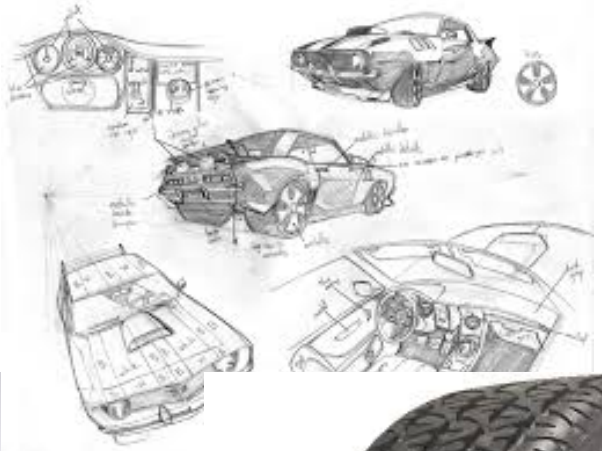
Science **328**, 1662 (2010)

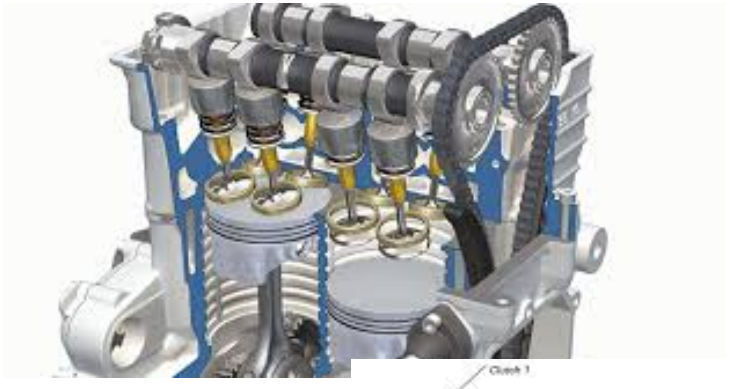
- Recreates structure and captures biophysics.
- Probes observed nanotoxicological response to nanoparticles (immune response), in particular in biophysical context (mimicking breathing).

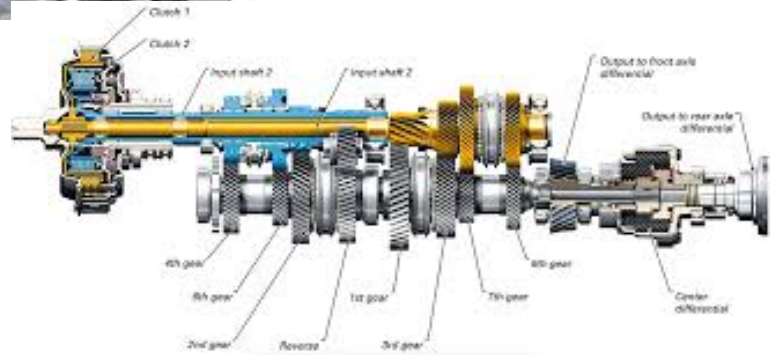
Higher Degree of Structure and Function in Tissues



An Engineering Approach

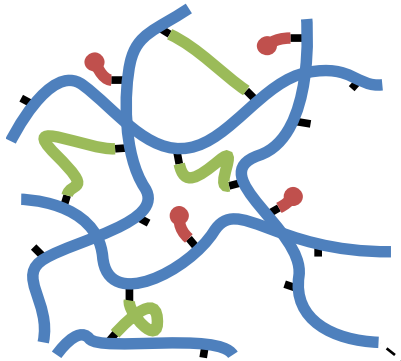




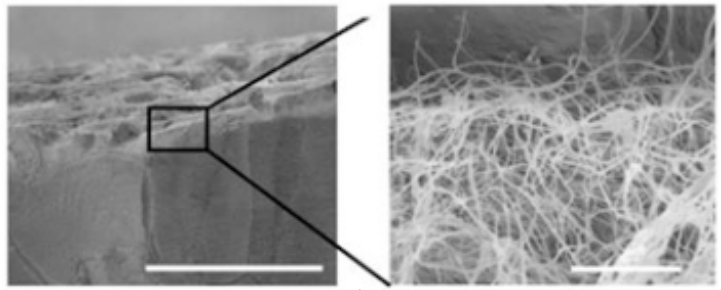


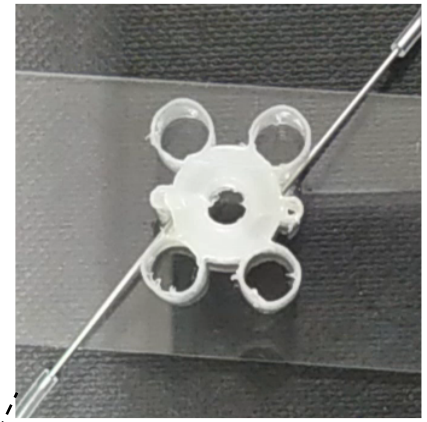
Multi-Scale Engineering of Compartmentalised Microvascularised Tissues

ECM Biochemistry
Physics/Mechanics
In situ gellation



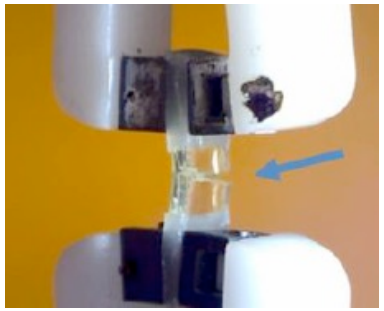
Biomimetic basement membrane

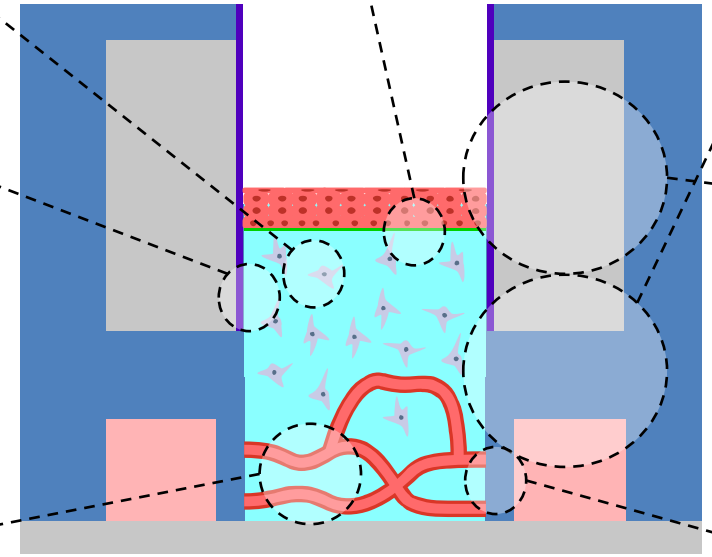


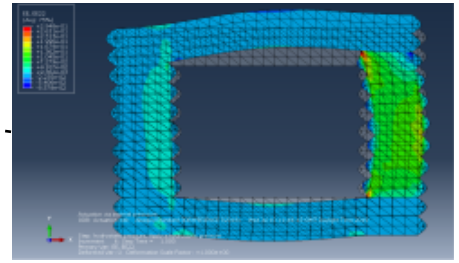


Compartmentalisation
Microfluidics
3D Printed

Bonding
Long-term structure

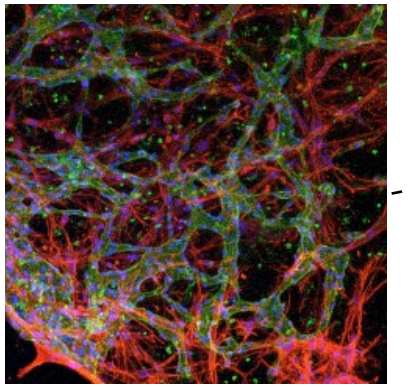


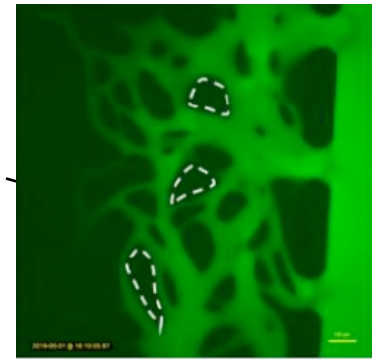




Biomechanics
Pneumatic

Stabilised co-culture





Perfusable vasculature

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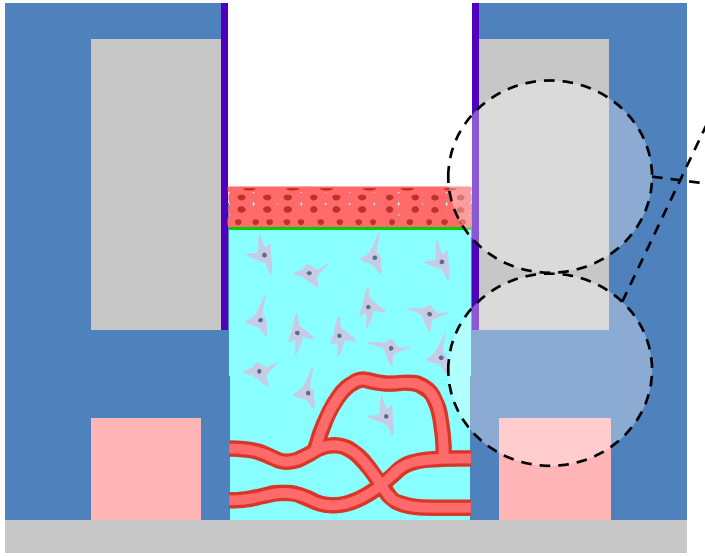
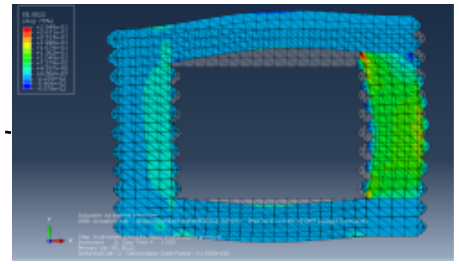
Microfabrication and Prototyping of Photocurable Silicones

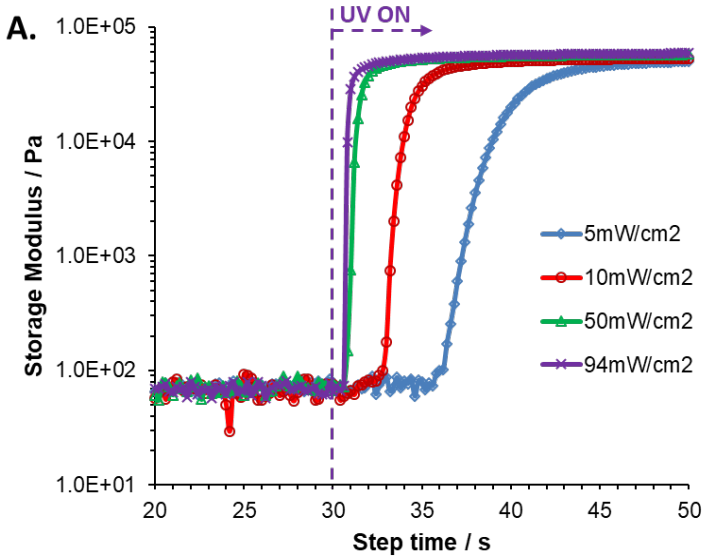
Compartmentalisation
Microfluidics
3D Printed

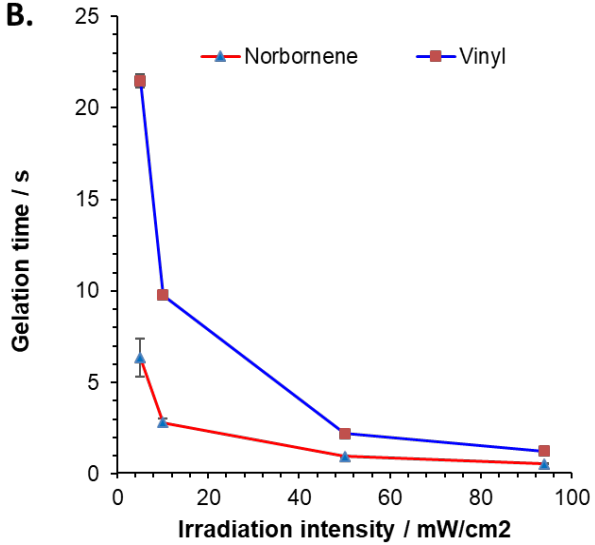


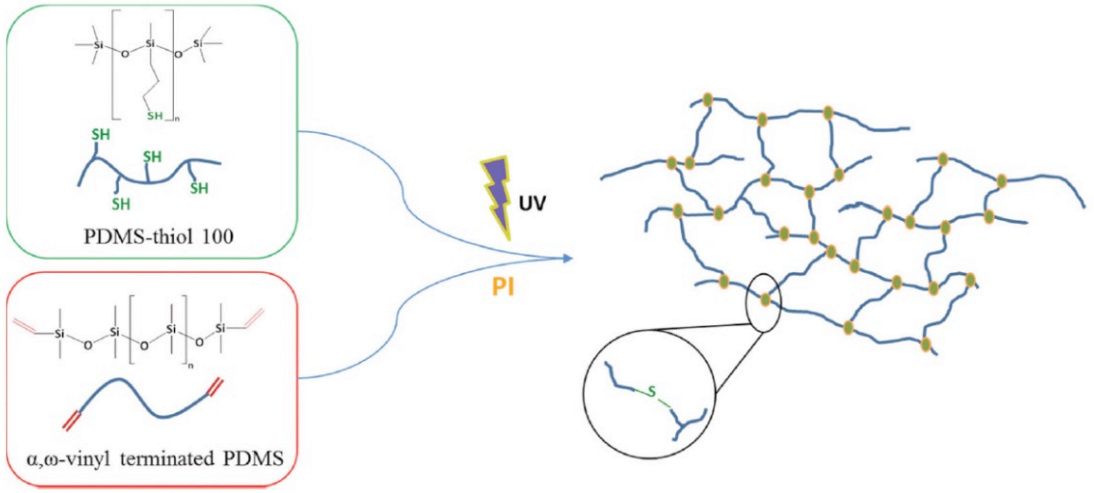


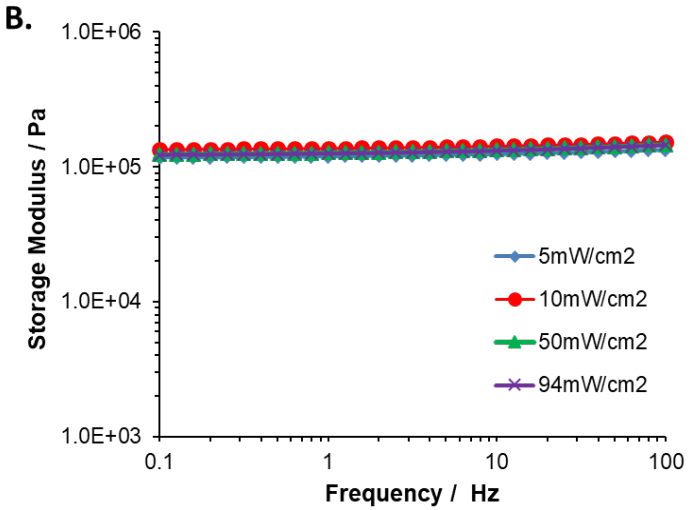
Biomechanics
Pneumatic

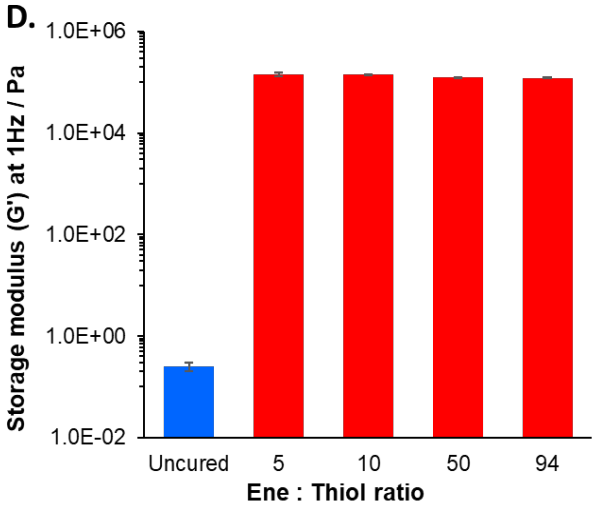
3D Ink Design – Ultra-Fast Cure Chemistry











- Ultra-fast cure chemistry.
- Cures in air without any oil residue.
- No toxic catalyst.

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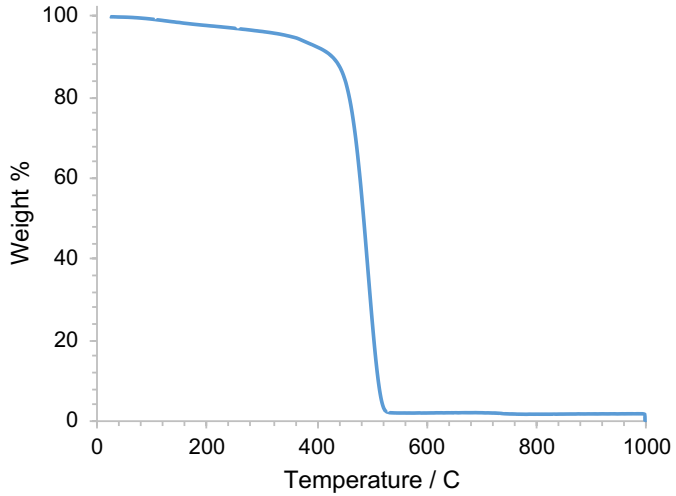
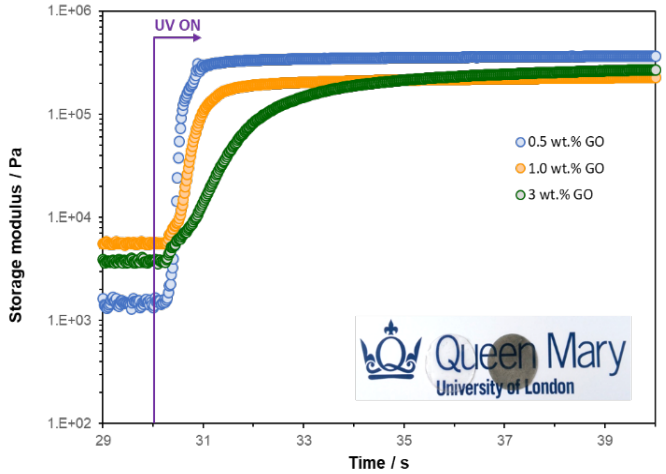
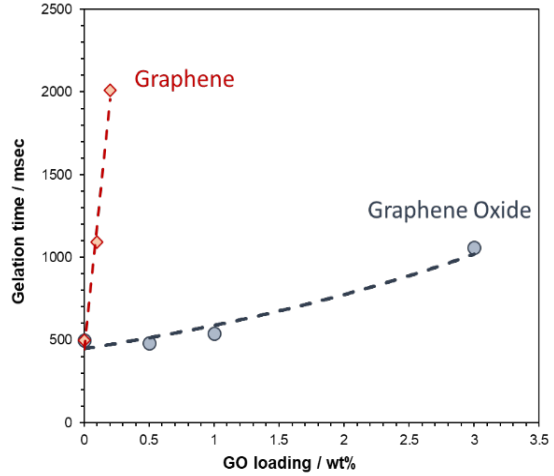
Graphene-Based Conductive Silicones

Excellent Thermal Stability



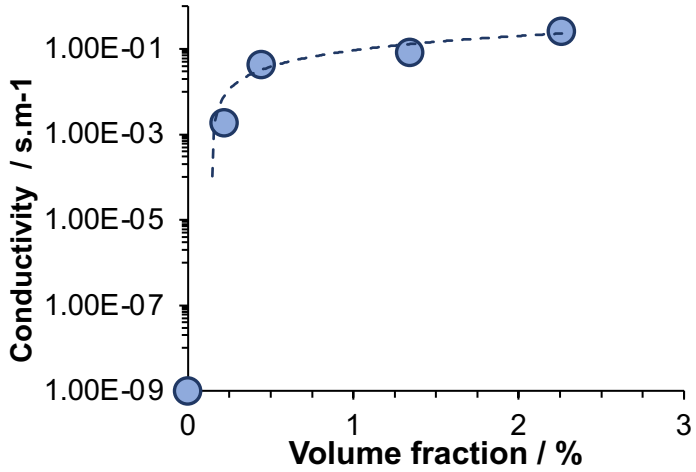
Fast Gelation of Opaque Composities





- Thermal stability up to 400°C
- Curing of composites with 3 wt% graphene oxide under 1s (250 μm samples).
- Achieve high conductivities even at 0.5 wt% after Gox thermal conversion.

Flowable, Printable Conductive Silicone Composites



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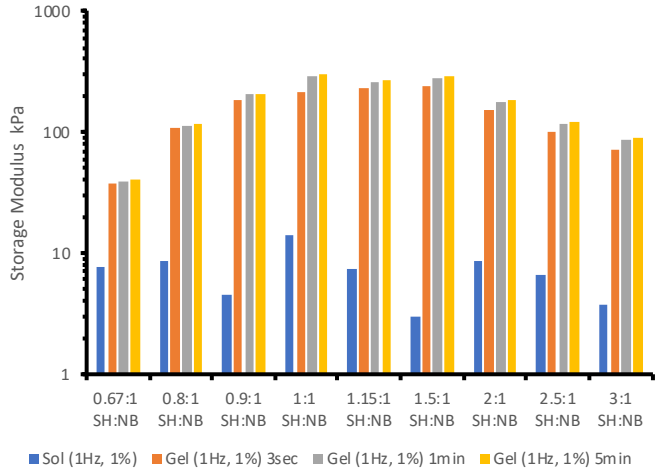
University of London

3D Printed Silicones – Flexible 3D Design for Organ-on-Chip Applications

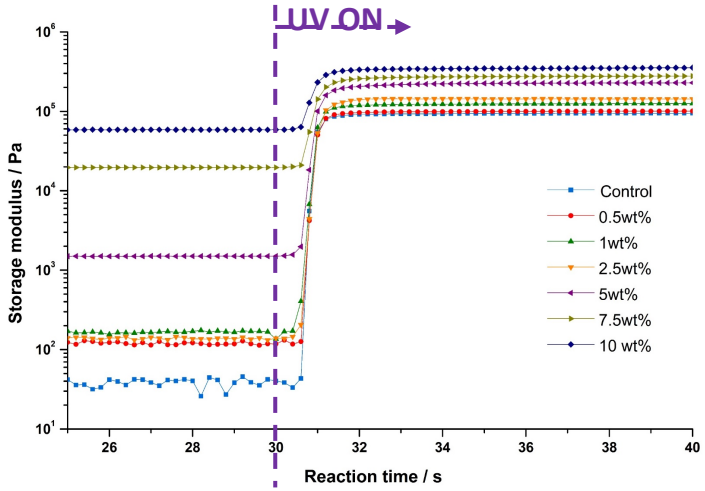
- Flexibility of design and automation.
- True 3D microstructuring.
- Limited resolution (> 100 microns).

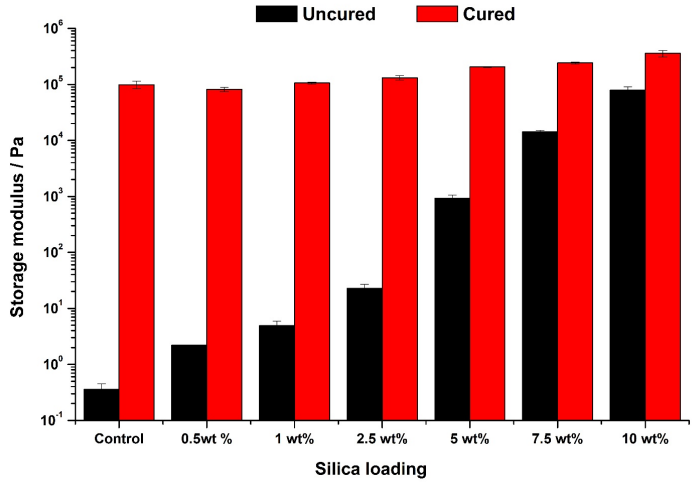
3D Ink Design - Formulation

Tuning Ink Mechanics



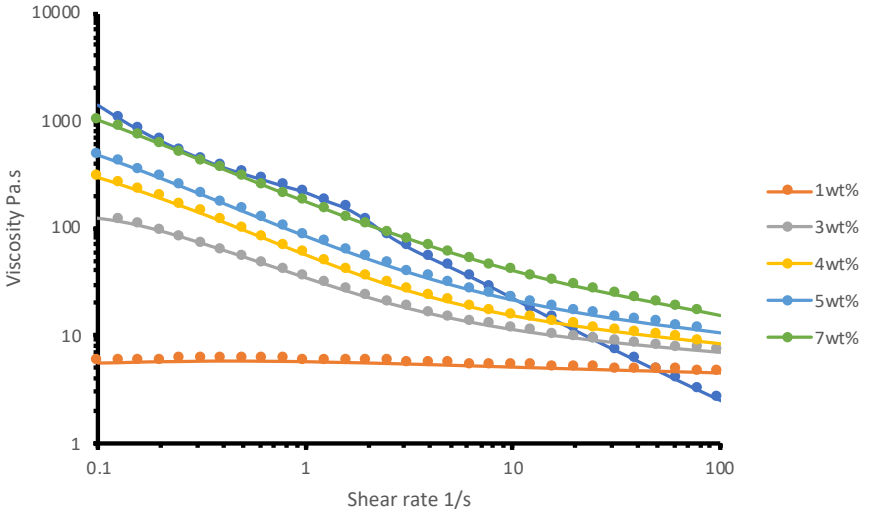
Rapid Curing of Filled Formulations



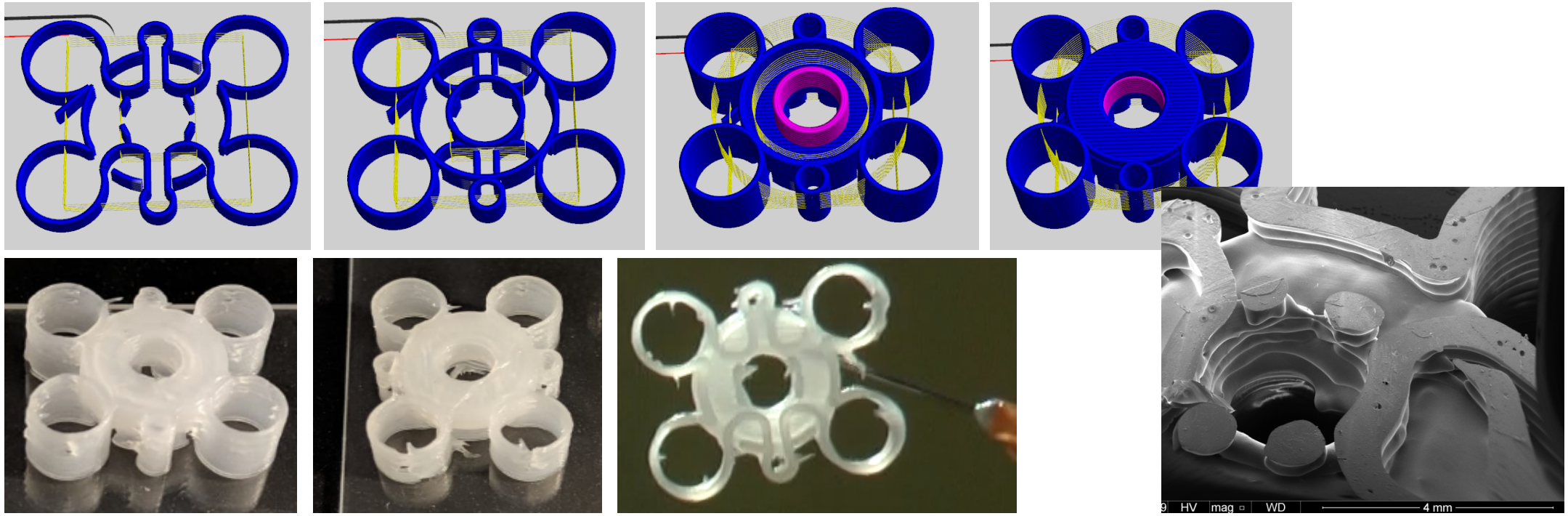


- Design of rheological properties of uncured formulations and mechanical properties of cured resins independently.
- Retention of fast curing even with opaque samples.
- Thixotropic properties suitable for extrusion-based printing.

Thixotropic Properties



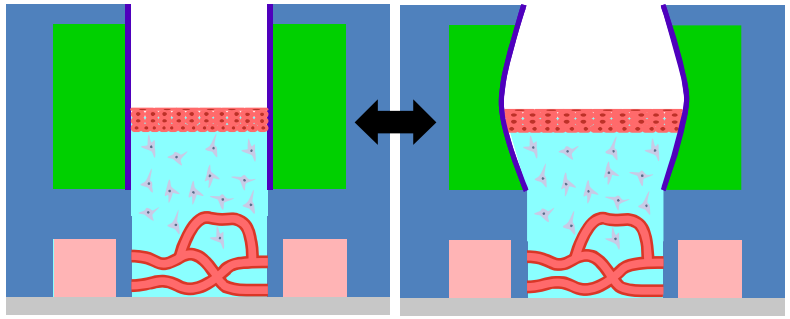
Chip Design and Printing



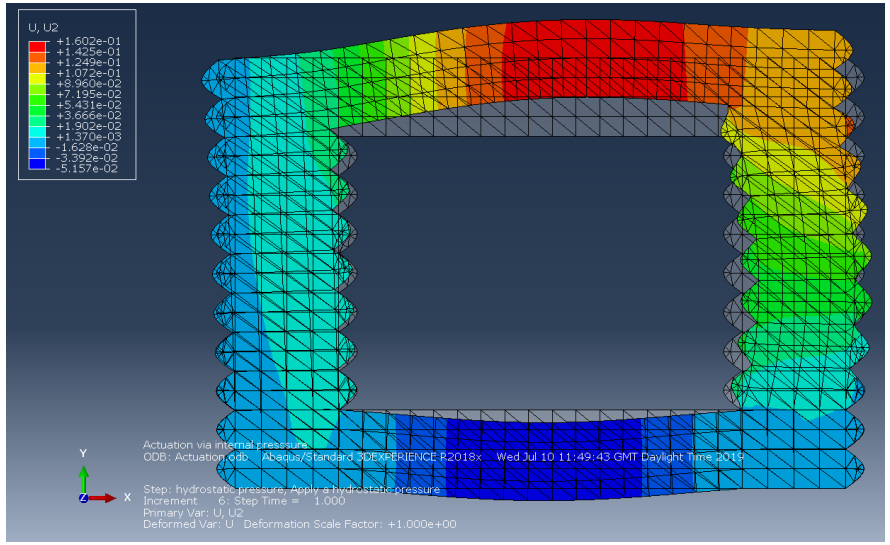
- More compact and symmetrical design to avoid uncontrolled nutrient/growth factor gradients.
- Introduce pneumatic chamber in upper compartment.

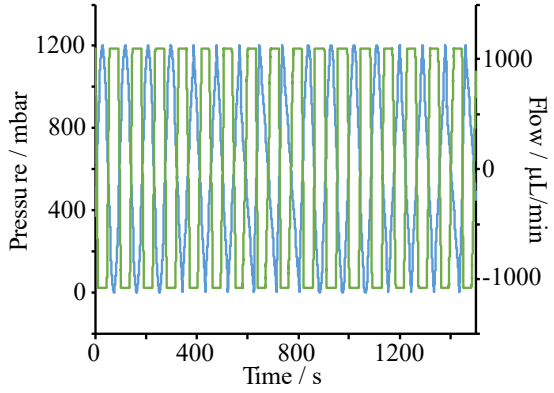
Pneumatic Chamber for Biomechanical Actuation

- Simple pneumatic chamber connected to software controlled pump.
- *In silico* prediction of the deformability of the compartment.



Predicting Deformation

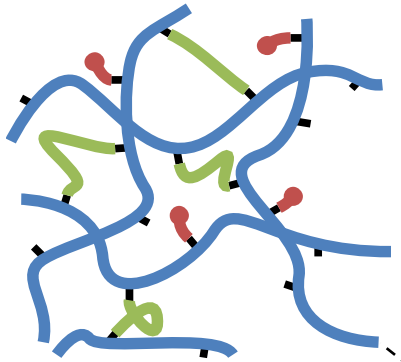




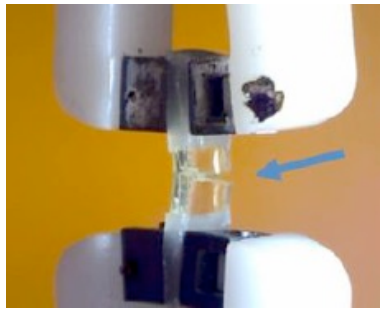
Cyclic Deformation

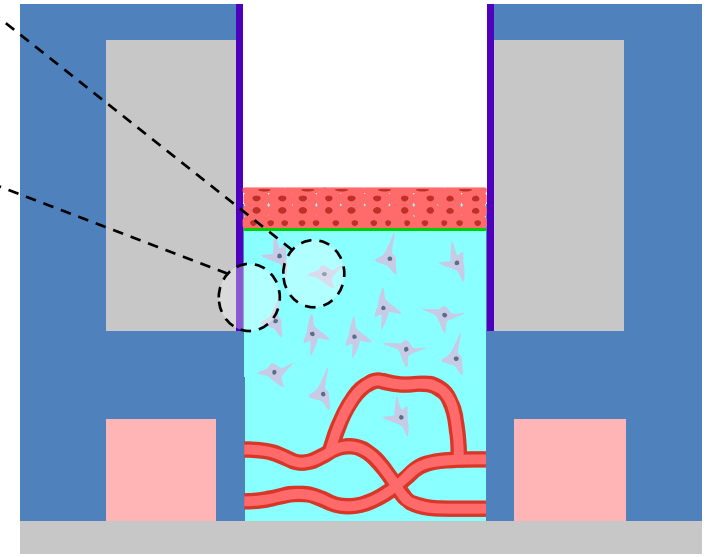
Mechanically Integrated Biomimetic Hydrogels

ECM Biochemistry
Physics/Mechanics
In situ gellation

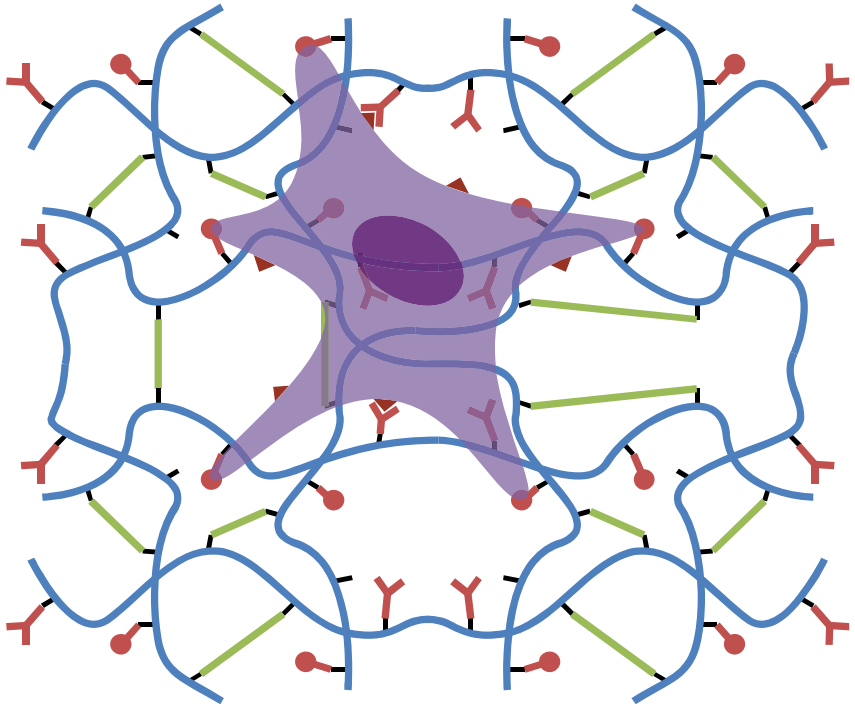


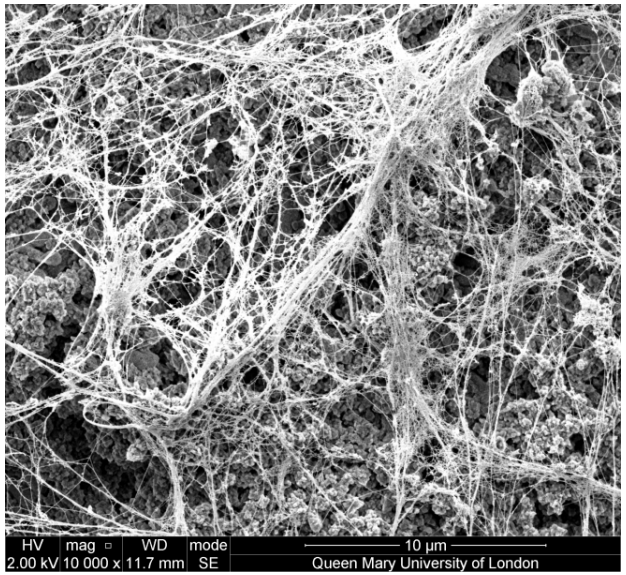
Bonding
Long-term structure

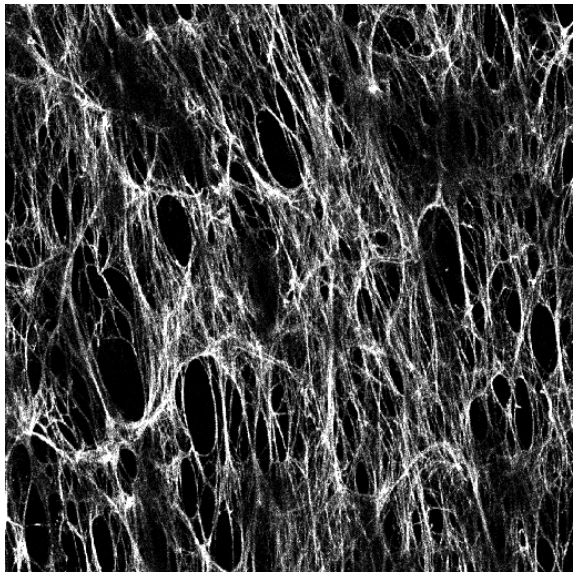




Mimicking the Cell Microenvironment

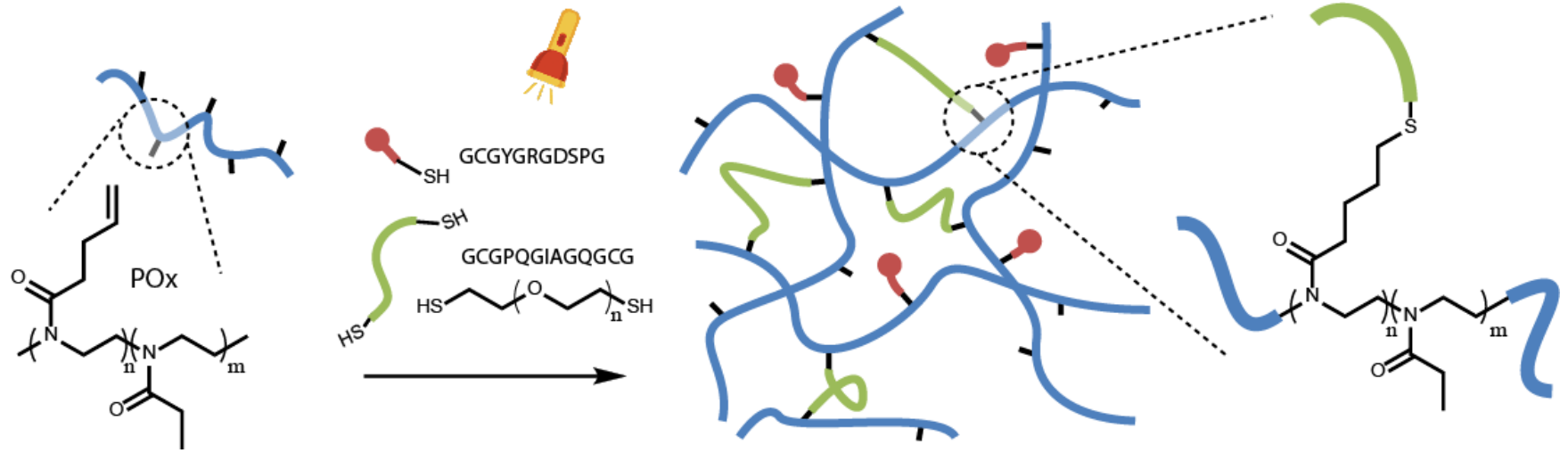






- Capture biochemical composition (cell adhesion, degradation, matrix deposition).
- Stiffness.
- Viscoelasticity.
- Porosity / morphology.

Peptide-Based Cell Degradable Hydrogels



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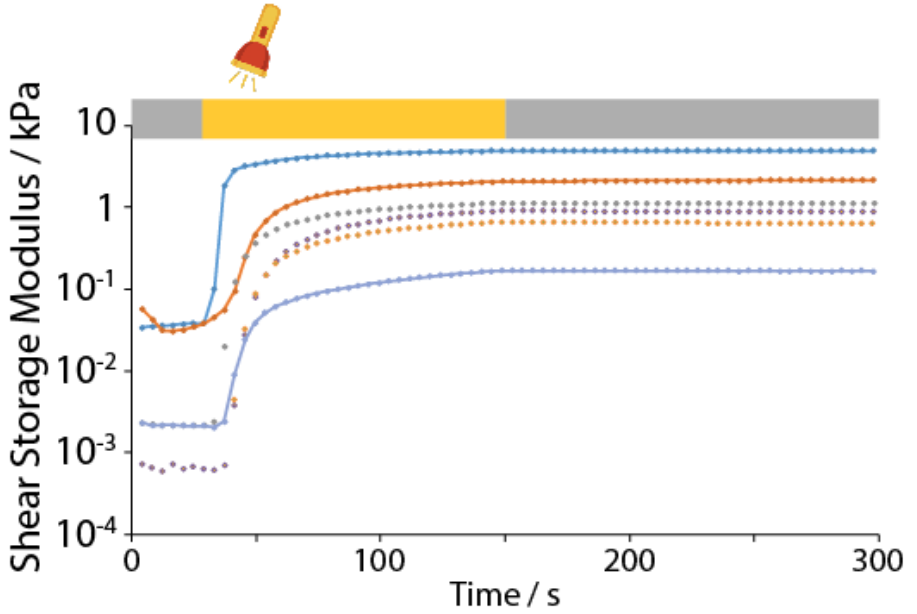
- Fast curing in physiological conditions.
- Control of mechanical properties and degradability.



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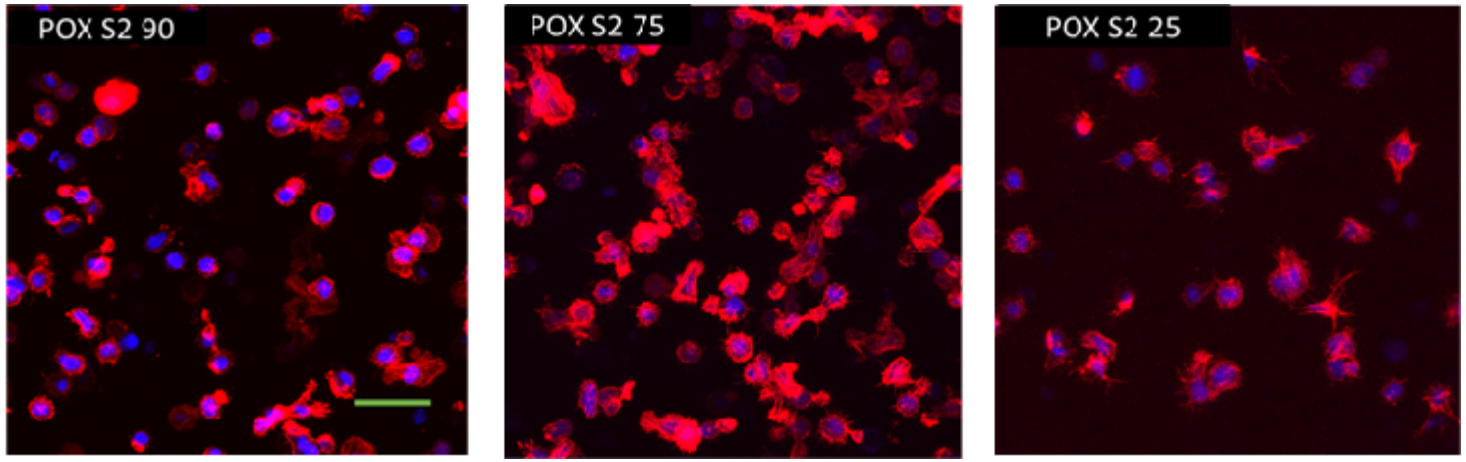
Regulation of Cell Spreading and Secretory Phenotype



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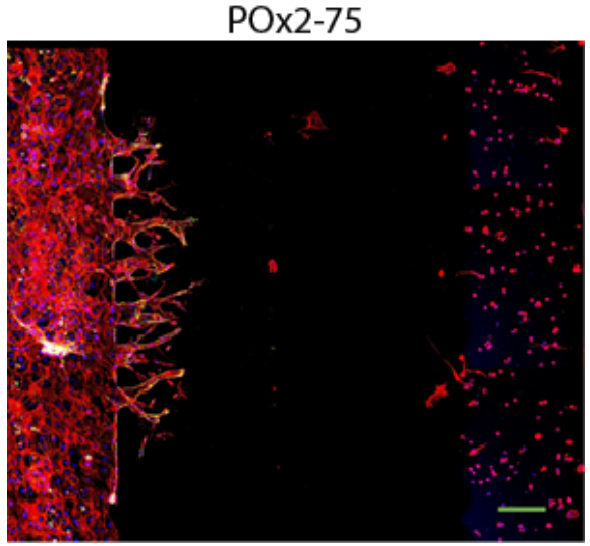
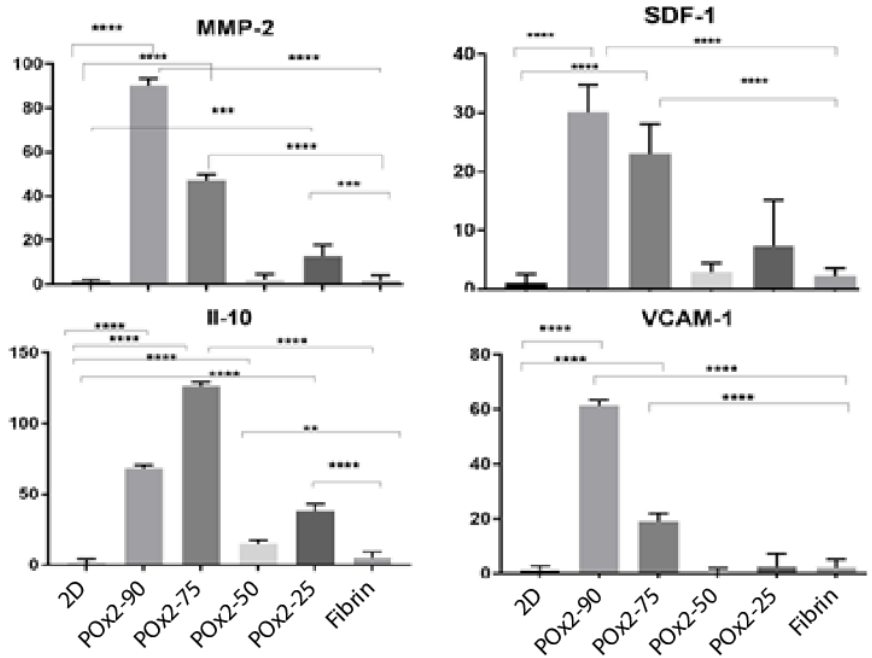
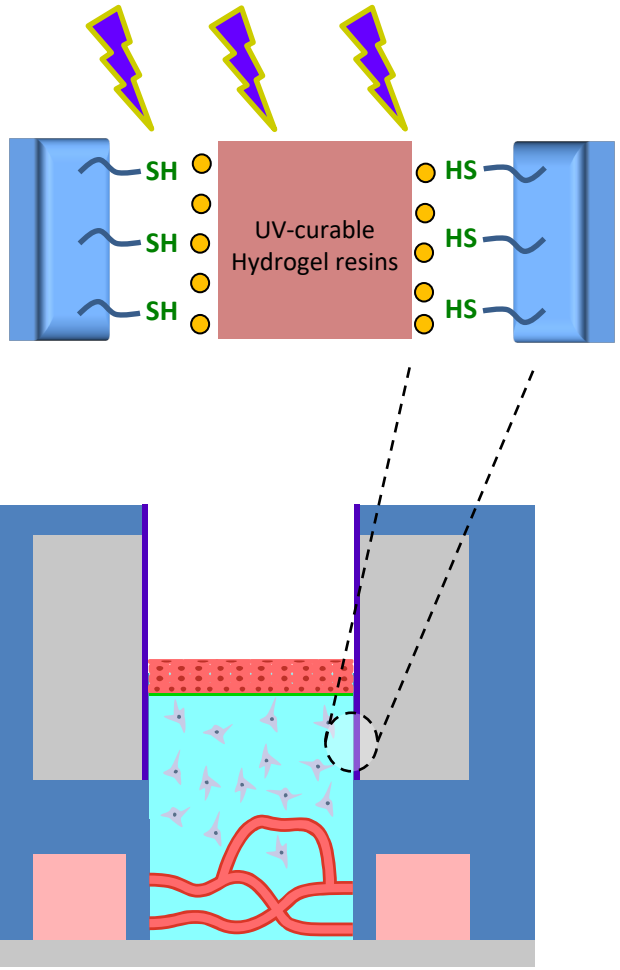


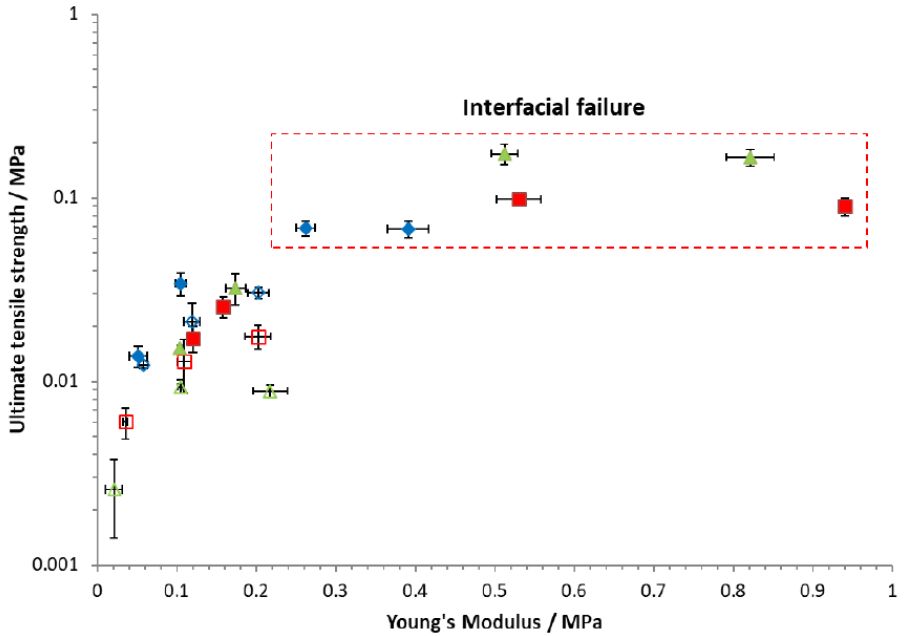


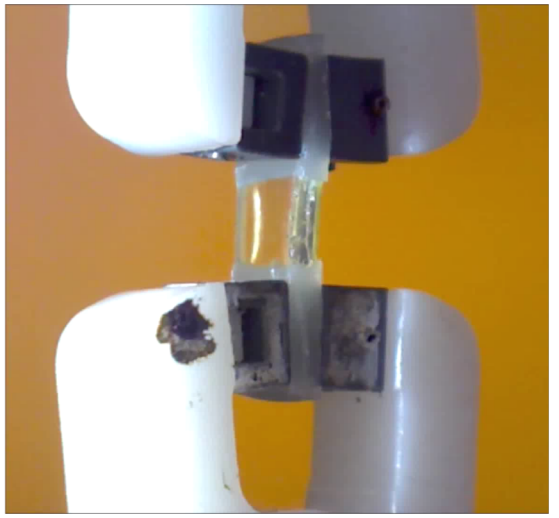
- Increased degradability correlates with cell spreading.
- Restriction of cell spreading correlates with increased growth factor secretion.
- Matrix engineering regulates pro-angiogenic phenotype.

Hydrogel Bonding in Thiol-Ene Hybrids



- Thiol-ene based hydrogels and silicones are chemically compatible.
- Improved adhesion compared to Sylgard PDMS.





Stretching rate : 1mm/min

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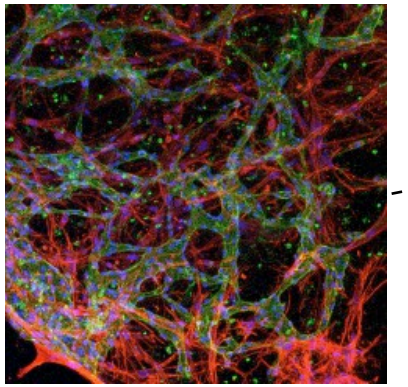
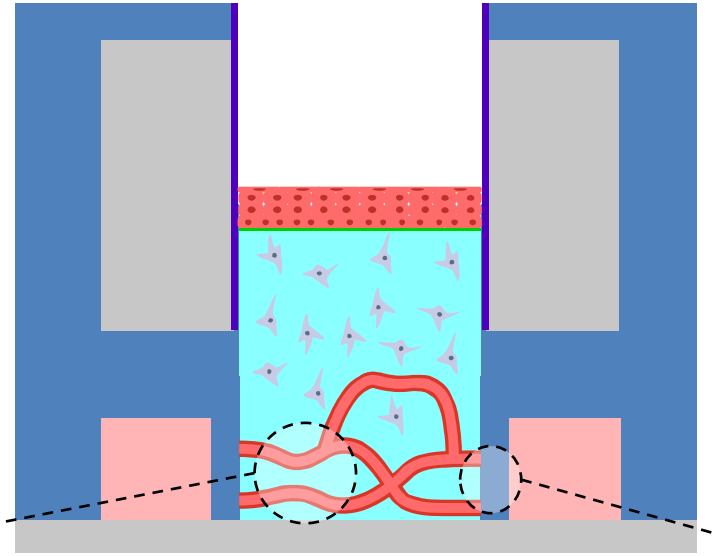
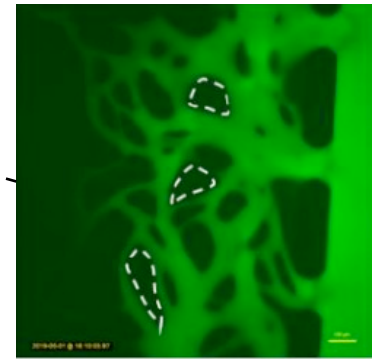
Engineering of Stable Perfusable Microvasculatures in Microfluidic Chips

Stabilised co-culture

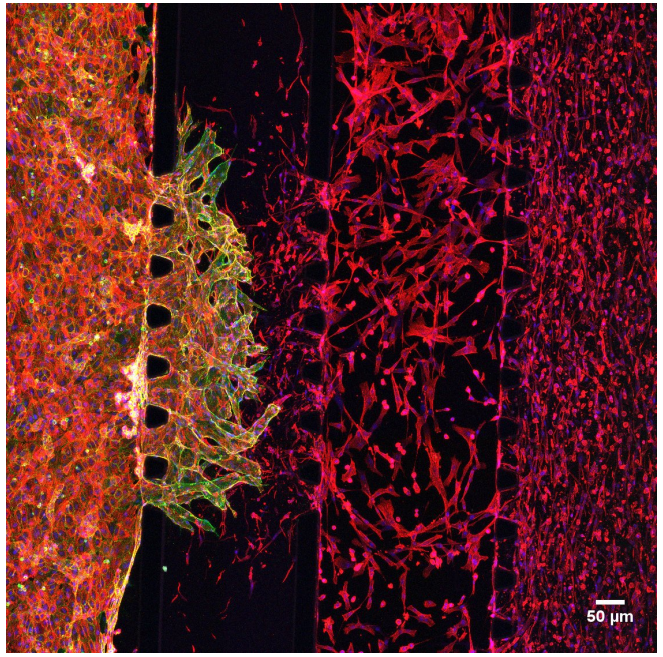


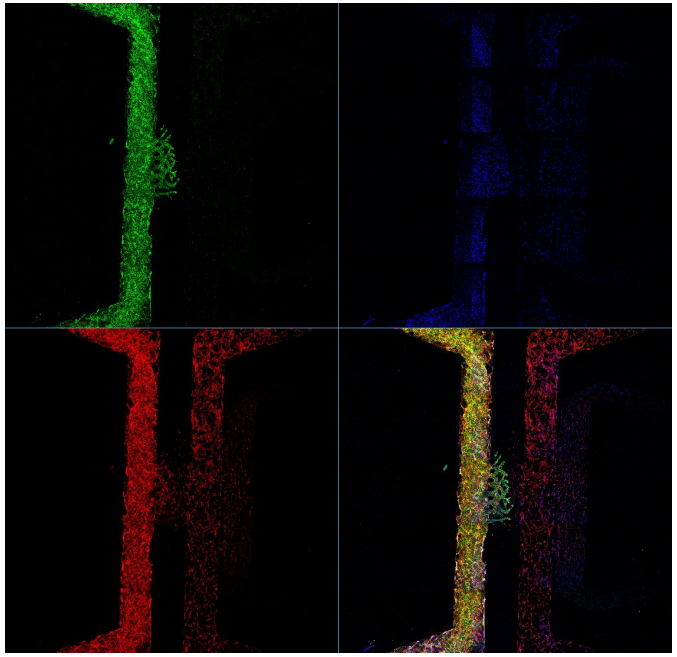




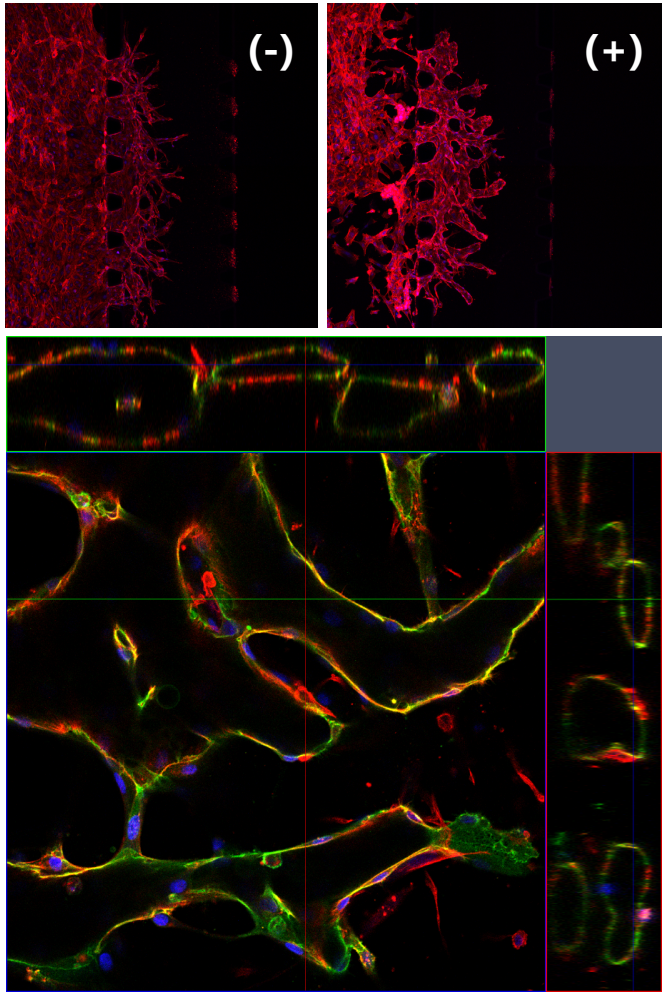
Perfusable vasculature

MSC Co-culture Promotes the Formatin of a Mature Microvasculature





F-Actin, CD31, DAPI



- Formation of perfusable microvascular network in chips.
- Interfacing with advanced multi-cellular in vitro models for safety/efficacy testing.

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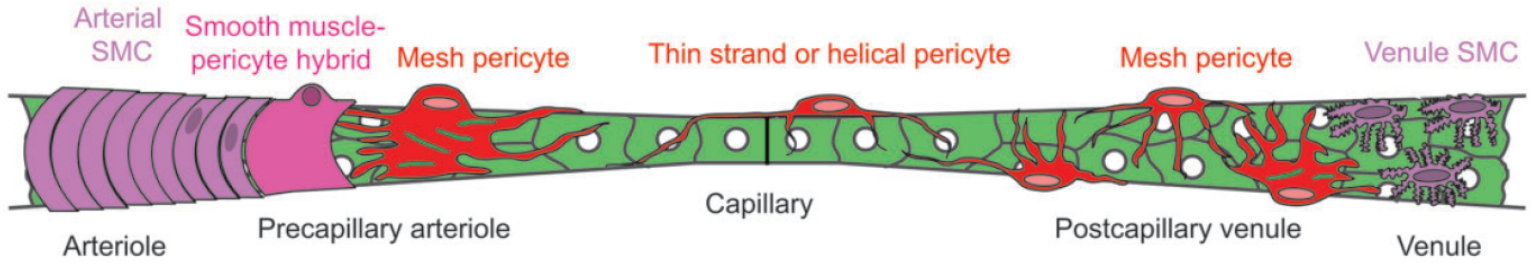
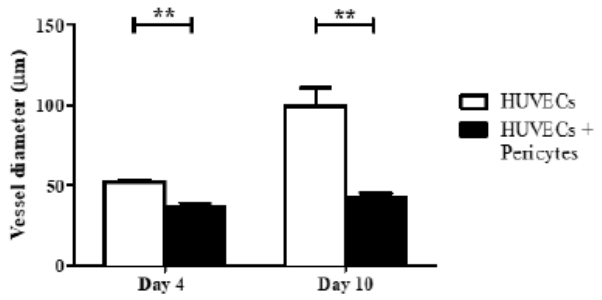
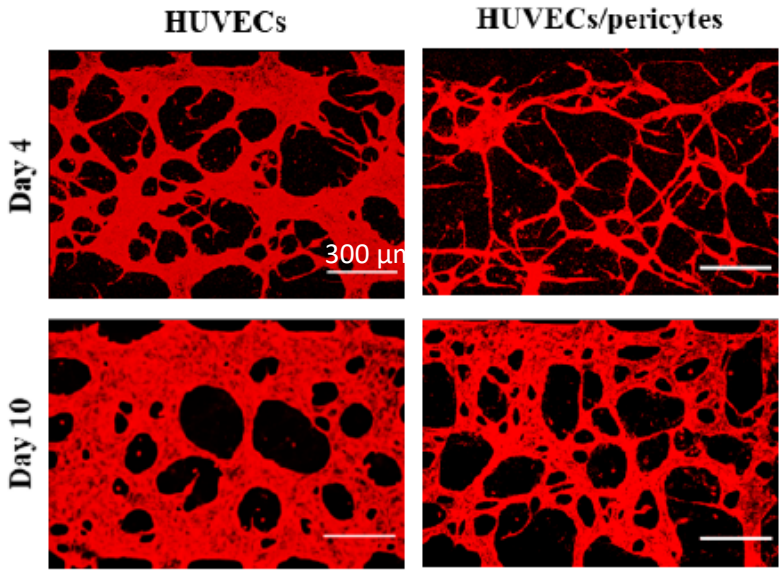
Pericyte Co-cultures for Stablisation of Microvasculatures

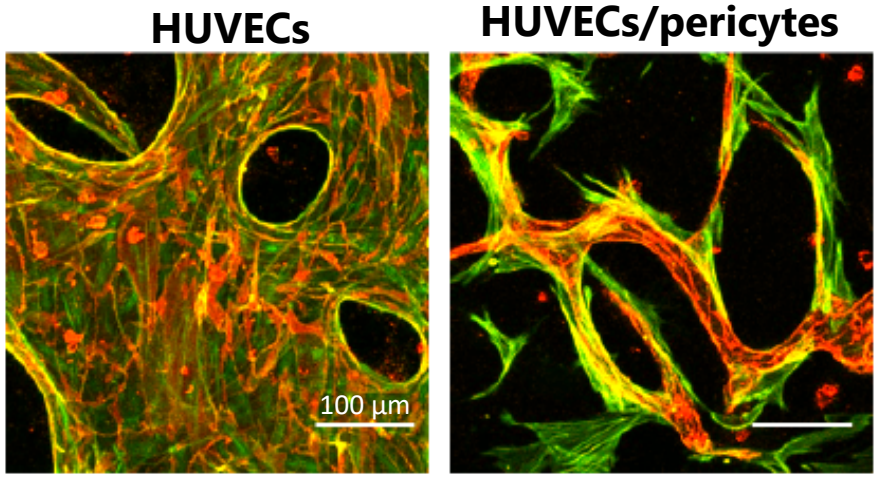


Attwell et al. Journal of Cerebral Blood Flow & Metabolism (2016).

Impact on Microvasculature Structure







CD31
Phalloidin

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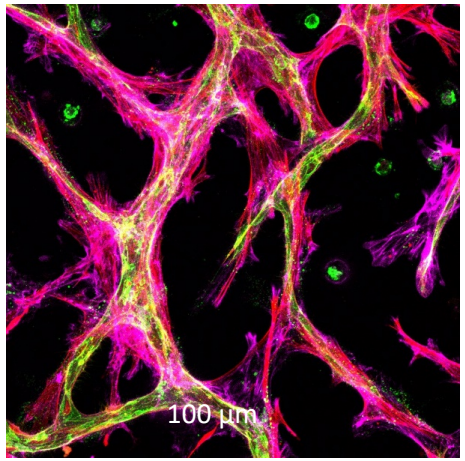
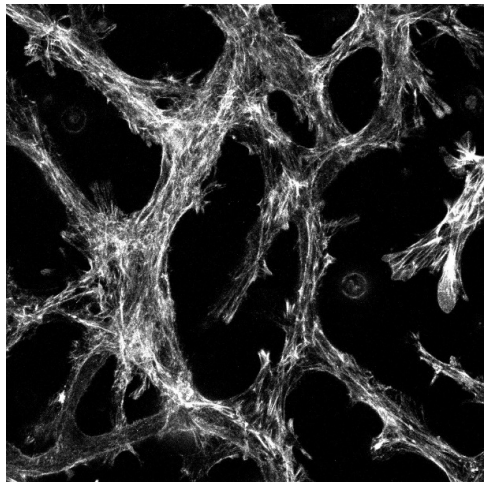
Long-Term Stability of Vascular Networks in Microfluidic Chips

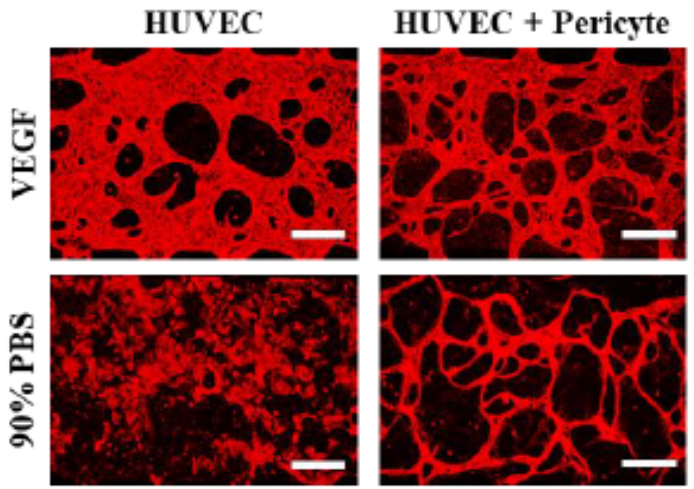
HUVECs + Pericytes

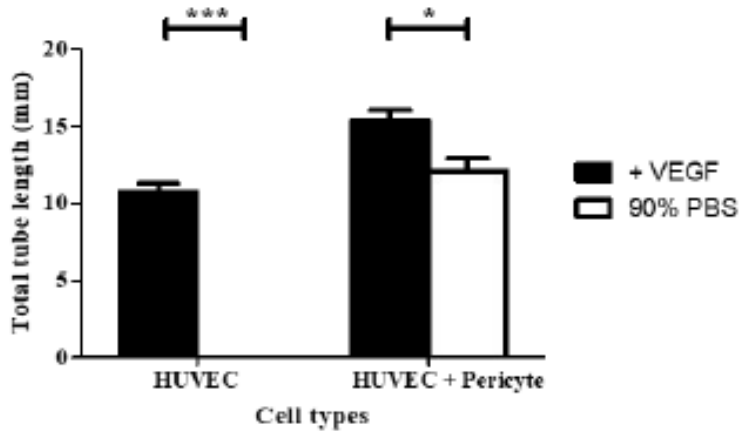


Fibronectin
CD31
F-actin



Fibronectin

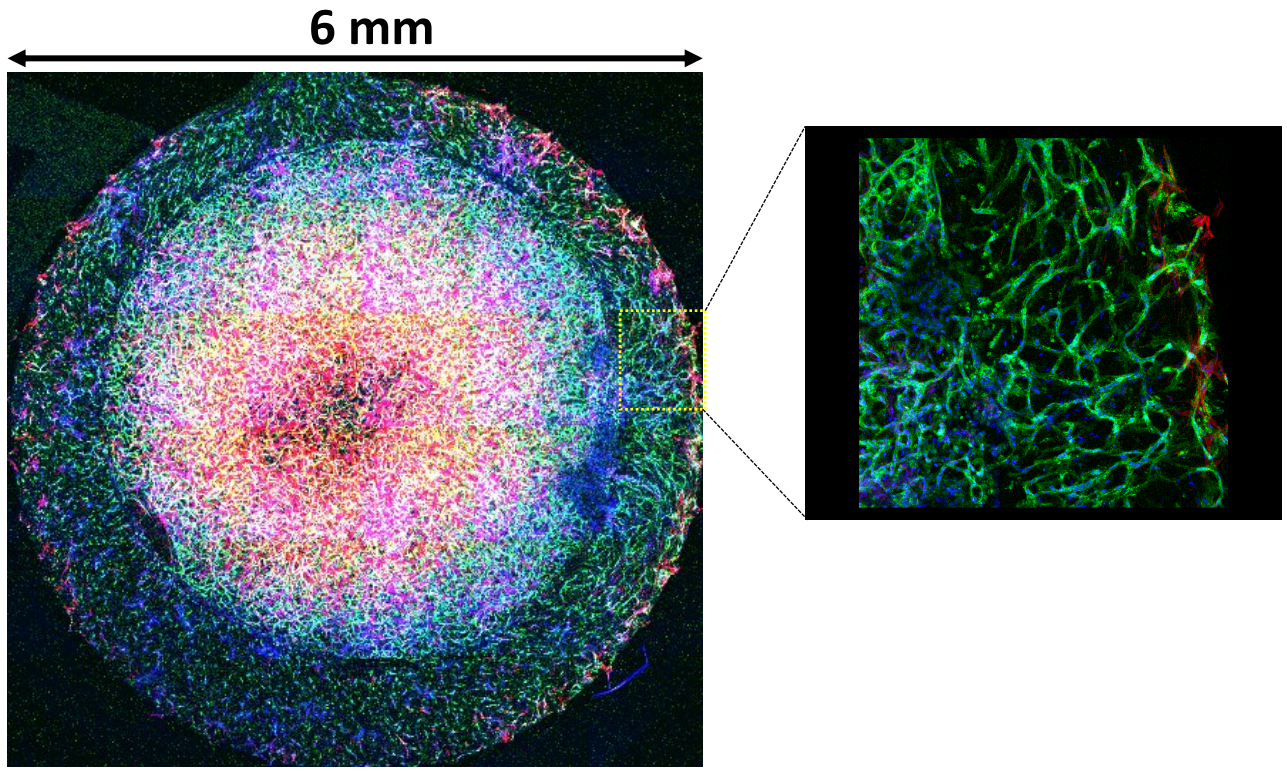




- Pericyte co-culture allows to stabilise microvascular networks for >3-4 weeks.
- Stable in multiple types of culture medium, even upon serum starvation.
- Compatible with implantation of more complex models.

Conclusions

- Thiol-ene PDMS display excellent properties for 3D printing in ambient conditions.
- Controlled mechanics and interfacing with hydrogels.
- Enables the fabrication of 3D chips for biomechanical actuation and the embedding of large complex multi-cellular tissue models.



Thank You

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