Week 8: Tissue Mechanobiology

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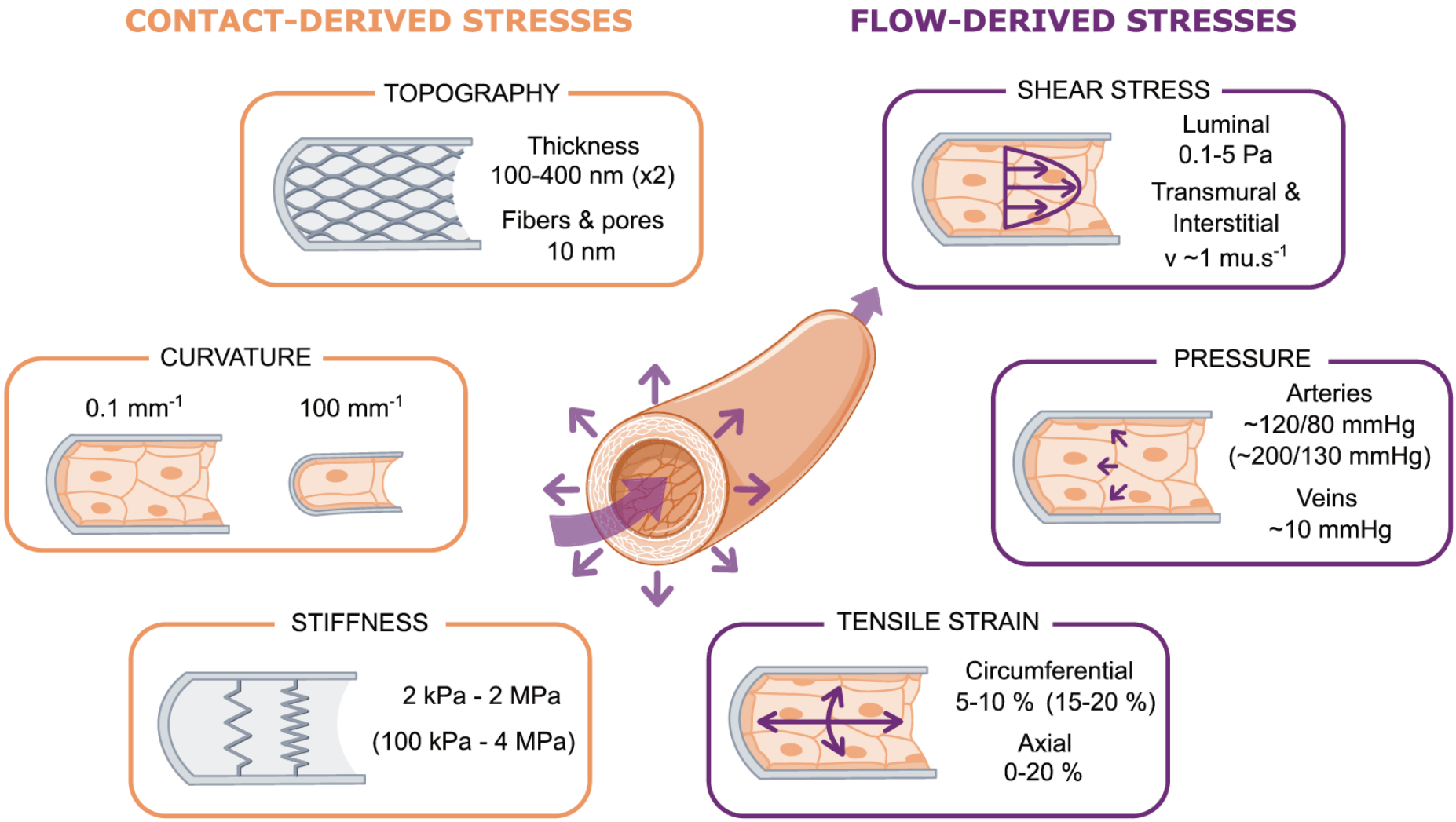
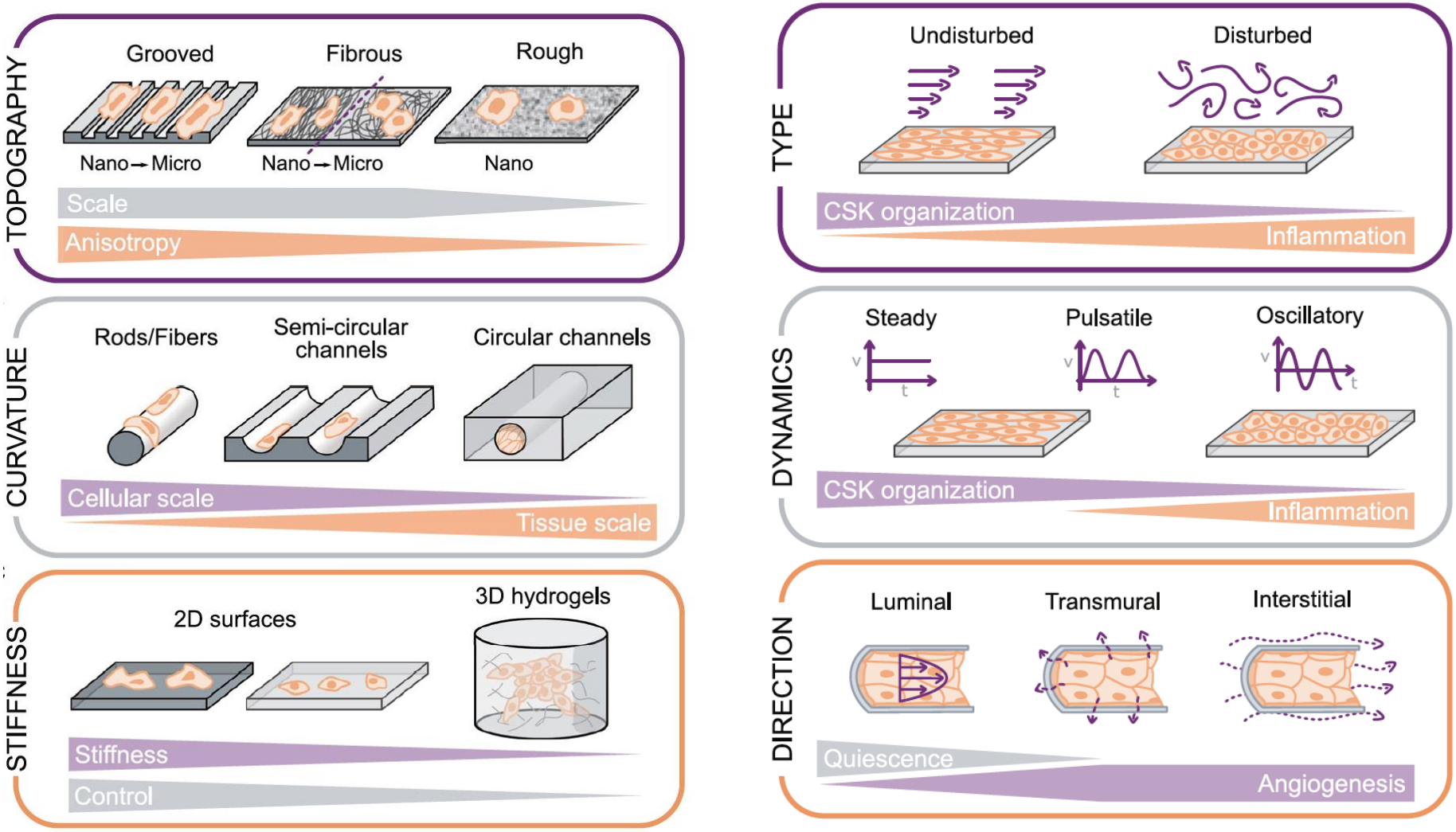
Endothelial Mechanobiology

- Endothelial cells cover the luminal surface of the vasculature
- A collection of intertwined stresses
- Contact stresses emanating from physical features of the underlying substrate
  - Substrate topography, curvature, stiffness
- Fluid-derived stresses due to blood flow
  - Shear (frictional) stress on the apical surface of endothelial cells, compressive blood pressure, circumferential (hoop) stresses

Endothelial Mechanobiology



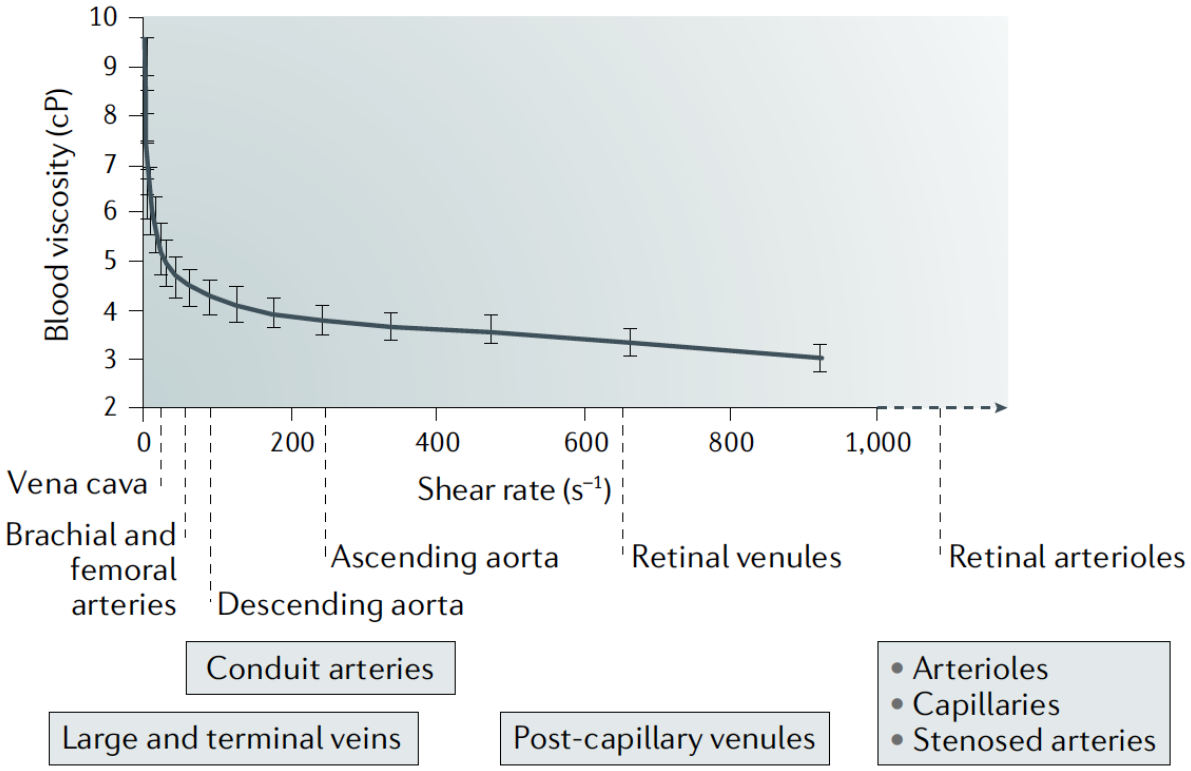
Endothelial Mechanobiology



Blood flow

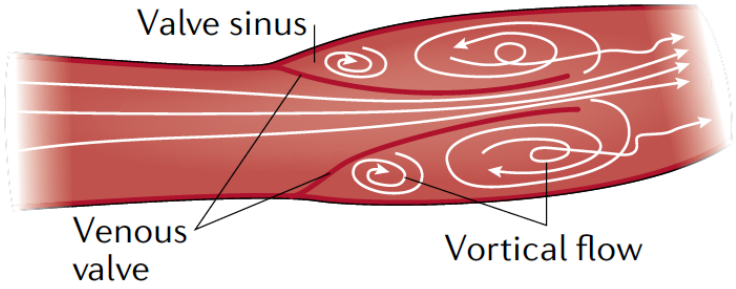
- Blood is a non-Newtonian fluid
- At sufficiently low shear rates (below 100s^{-1}) exhibits shear-thinning

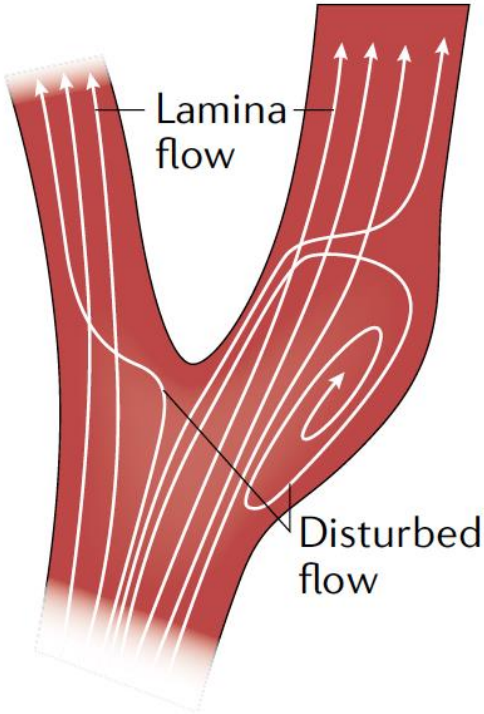
- Time-averaged wall shear stress: 1Pa (aorta) to 5 Pa (arterioles)



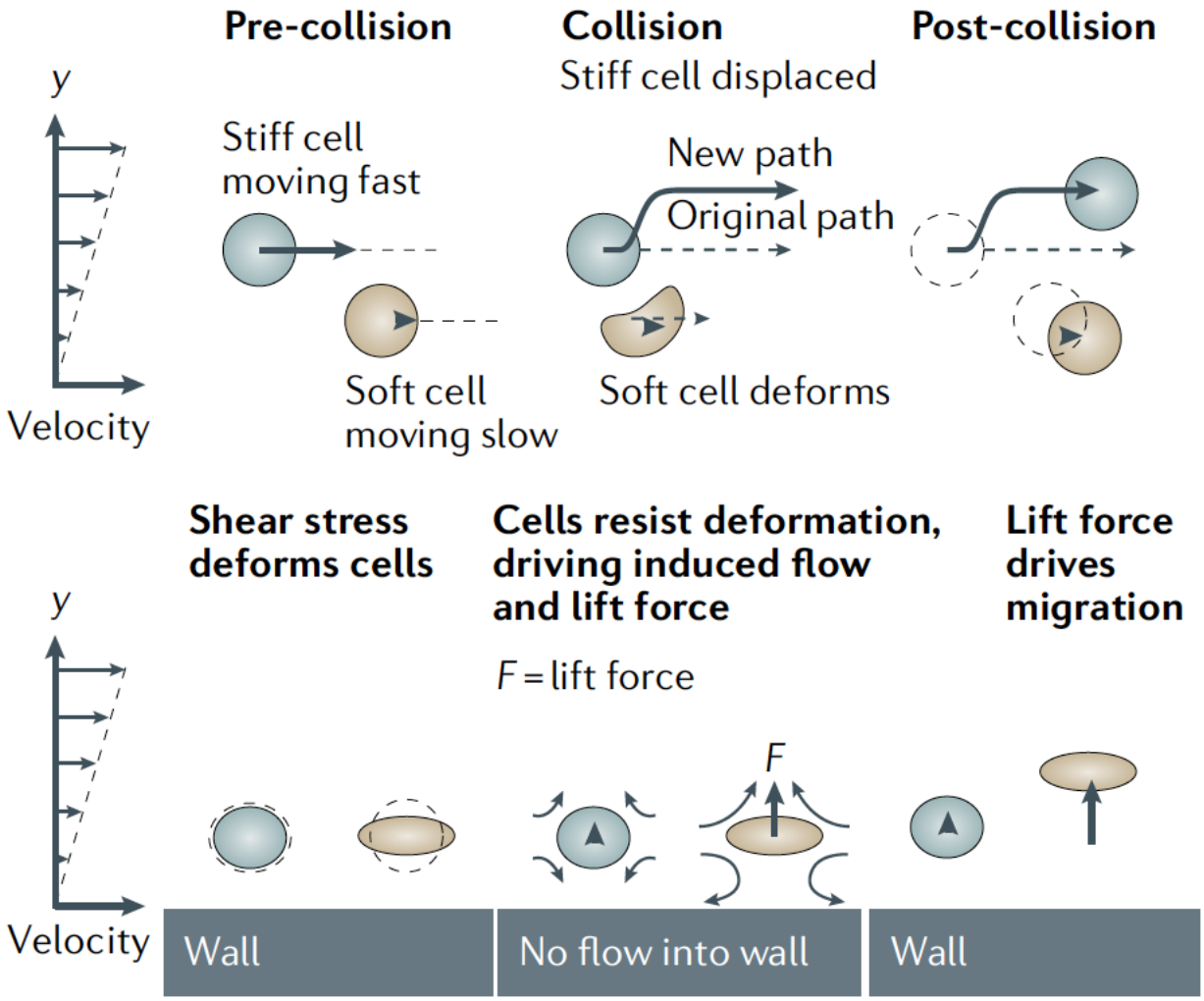
Blood flow

- **Undisturbed flow** is laminar flow
- **Disturbed flow**: turbulent or laminar flow with spatial shear stress gradient and vortices
  - Vascular curvature, branching, bifurcations
  - Many vascular diseases occur in these regions: atherosclerosis, thrombosis, etc.





Blood cells


Pre-collision
Collision
Stiff cell displaced
Post-collision
y
Velocity
Stiff cell moving fast
New path
Original path
Soft cell moving slow
Soft cell deforms
Shear stress deforms cells
Cells resist deformation, driving induced flow and lift force
Lift force drives migration
F = lift force
F
Wall
No flow into wall
Wall

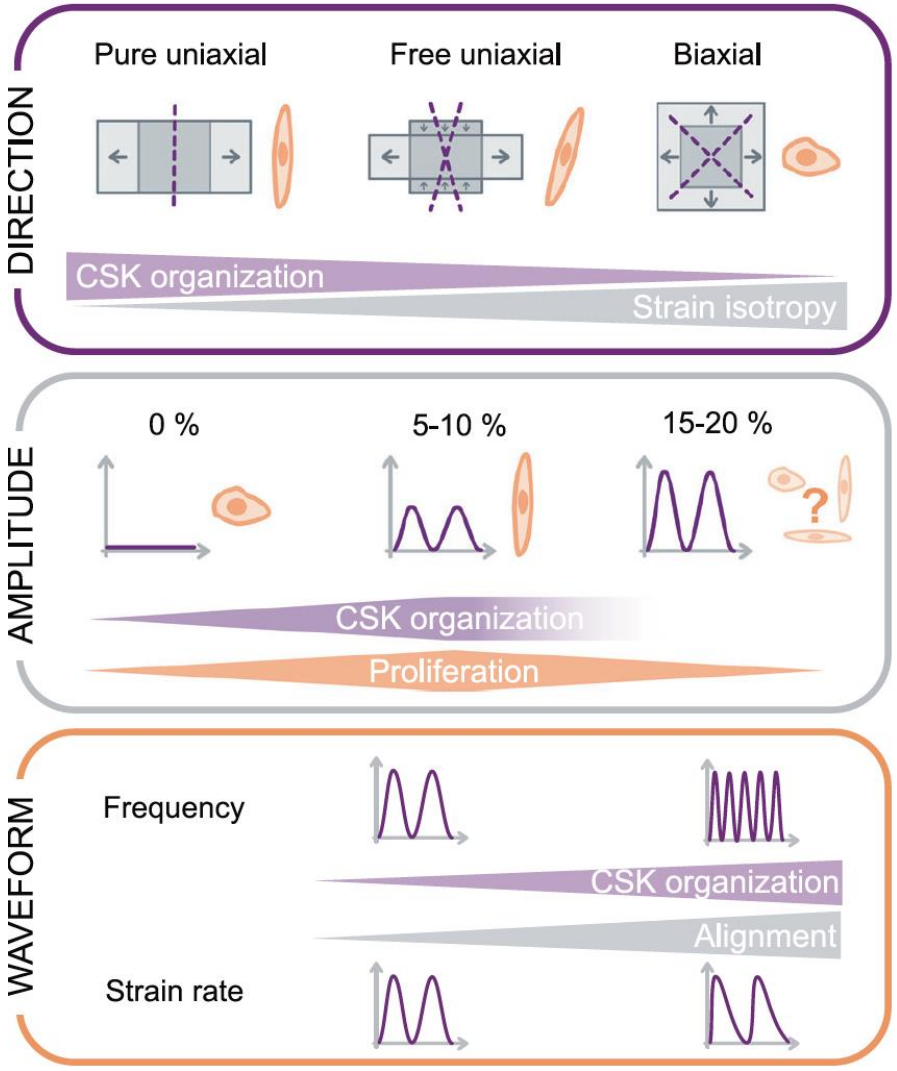
Pressure and stresses

- Blood pressure varies along the vasculature
  - From 1.3 kPa (veins) to 16 kPa (aorta)
  - Hypertension: 27 kPa
  - Pulsatile on the arterial side, dampened in capillaries

- Alignment, elongation, cytoskeletal reorganization, proliferation

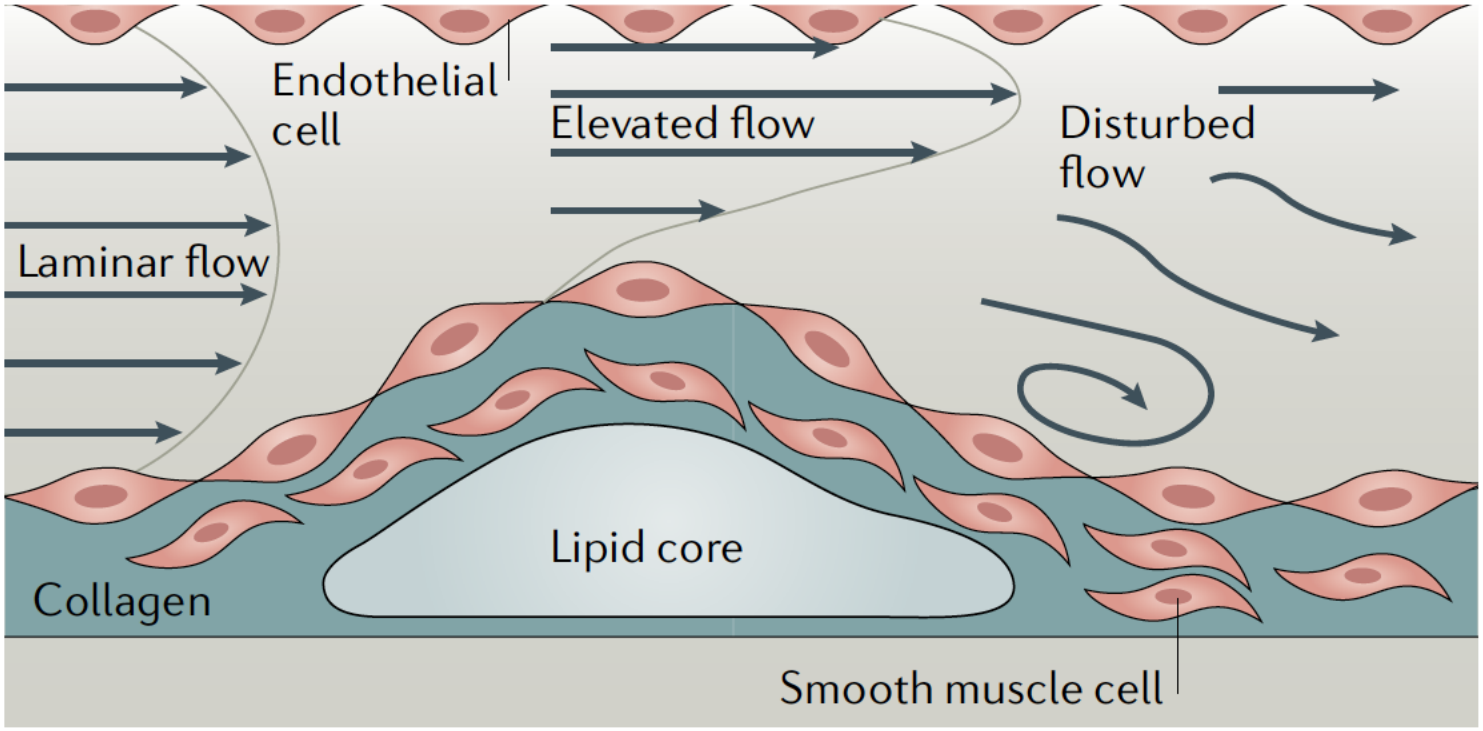
- Axial tensile stresses: tissue growth or movement: up to 20% strain
- Circumferential stresses: transmural pressure difference that dilates the vessels cyclically: up to 15% strain
- Physiological range: 5-10% strain

Features of tensile stresses

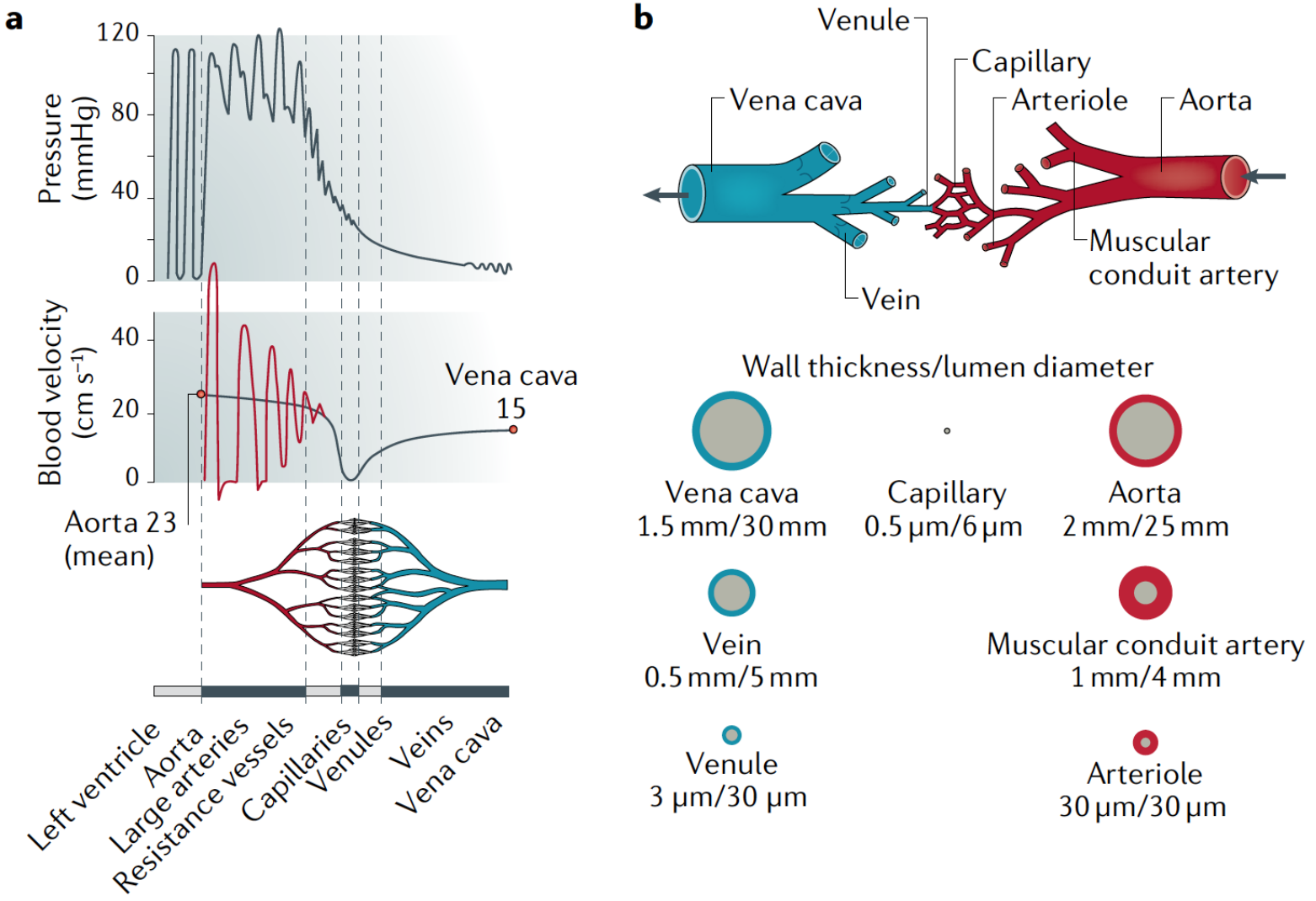


How do endothelial cells sense forces?

- Mechanosensitive ion channels
- Primary cilia
- Cadherins and integrins
- Notch complex



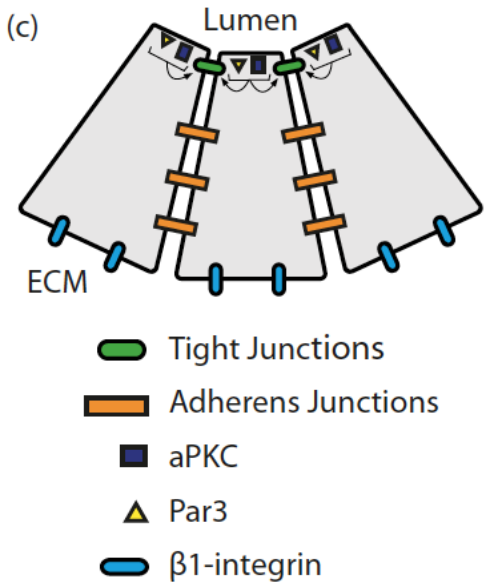
Anatomy and mechanical homeostasis

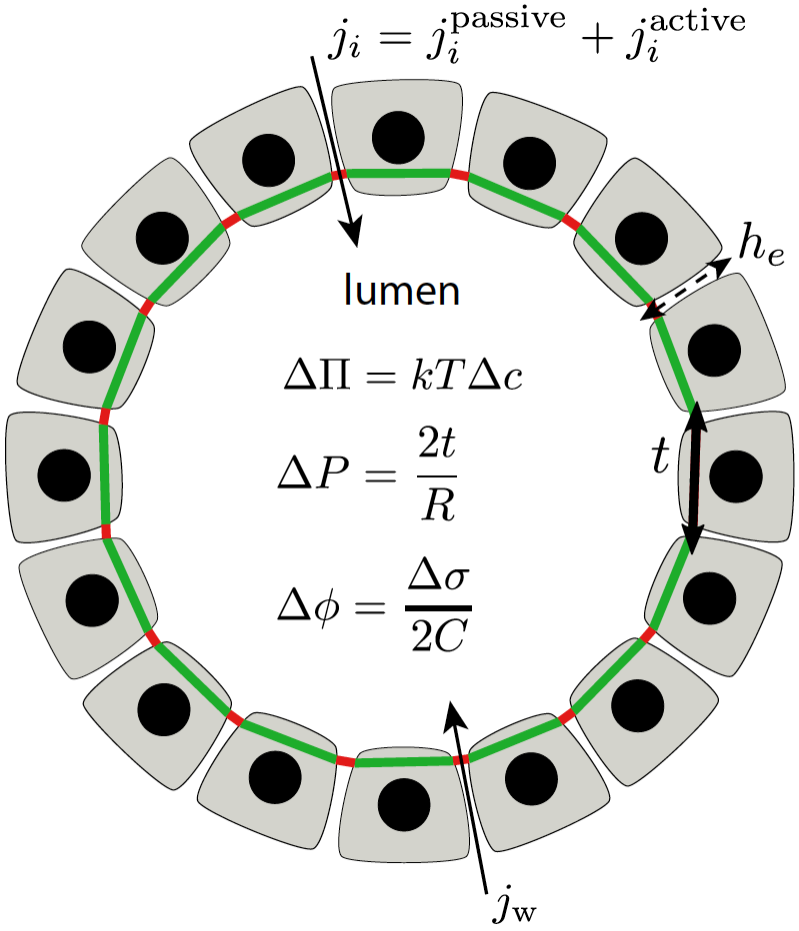


Anatomy and mechanical homeostasis

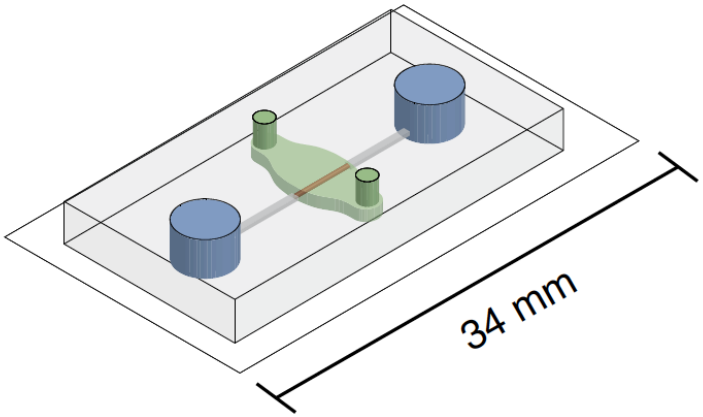
- Osmotic pressure
- Hydrostatic pressure
- Electrostatic potential

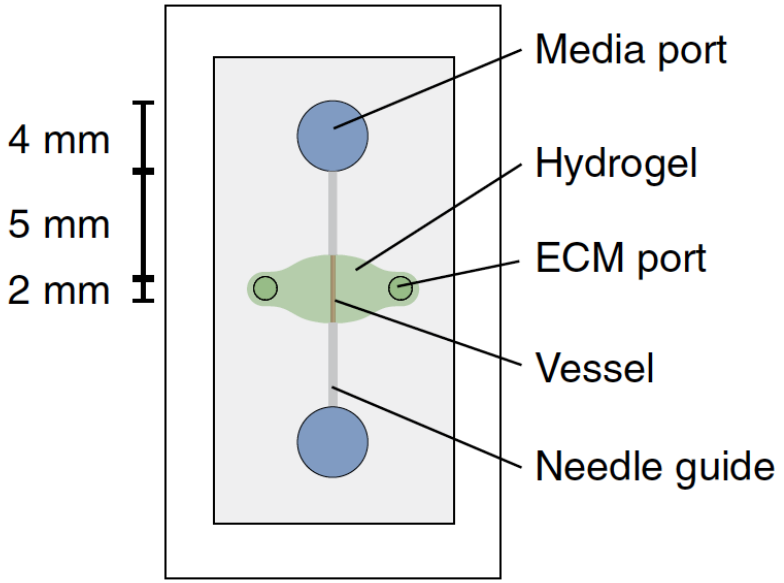
- Laplace Law and tension





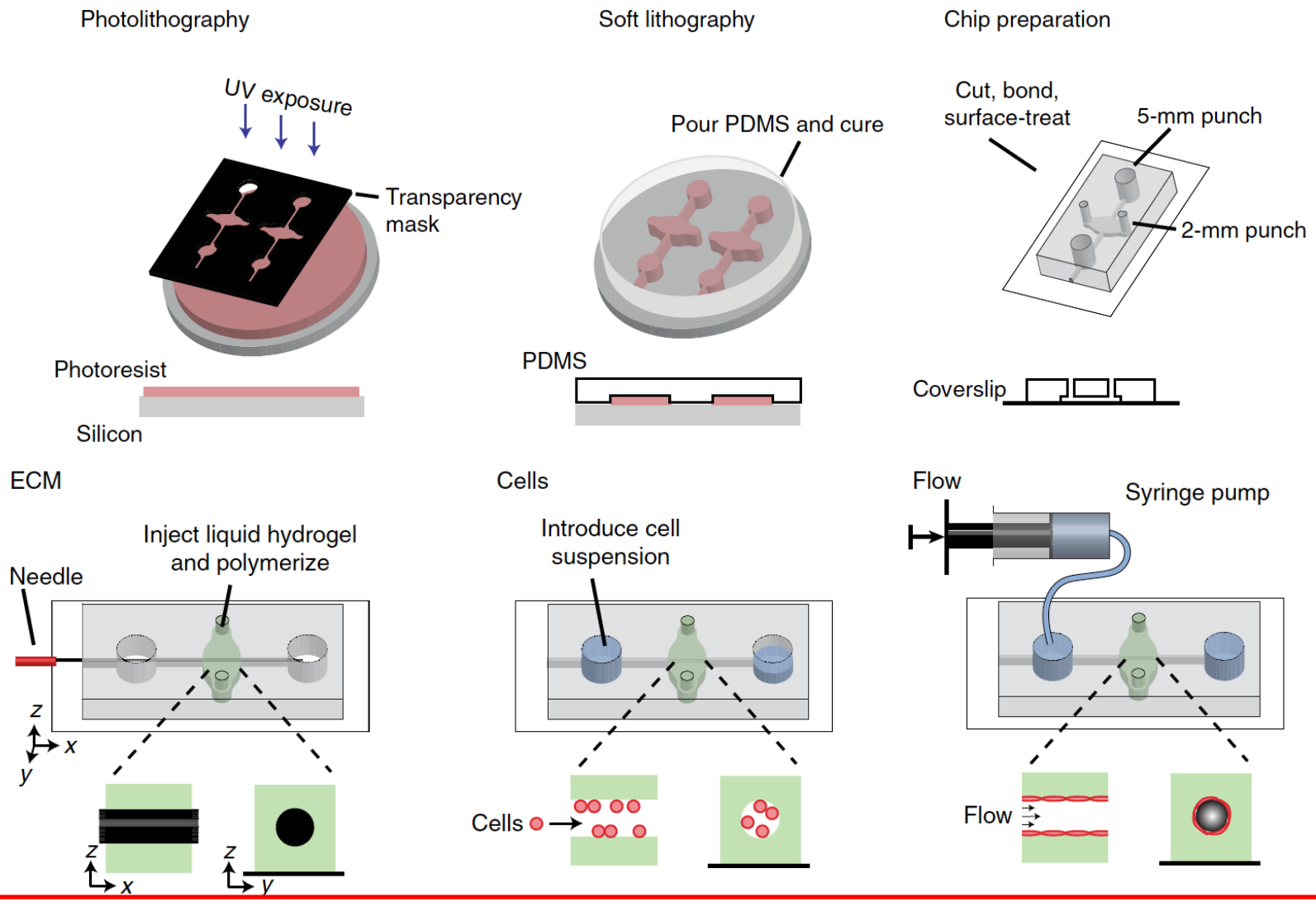
Microfabricated blood vessels





$$Q = \frac{\pi r^3}{4\mu}\tau_0$$

Microfabricated blood vessels



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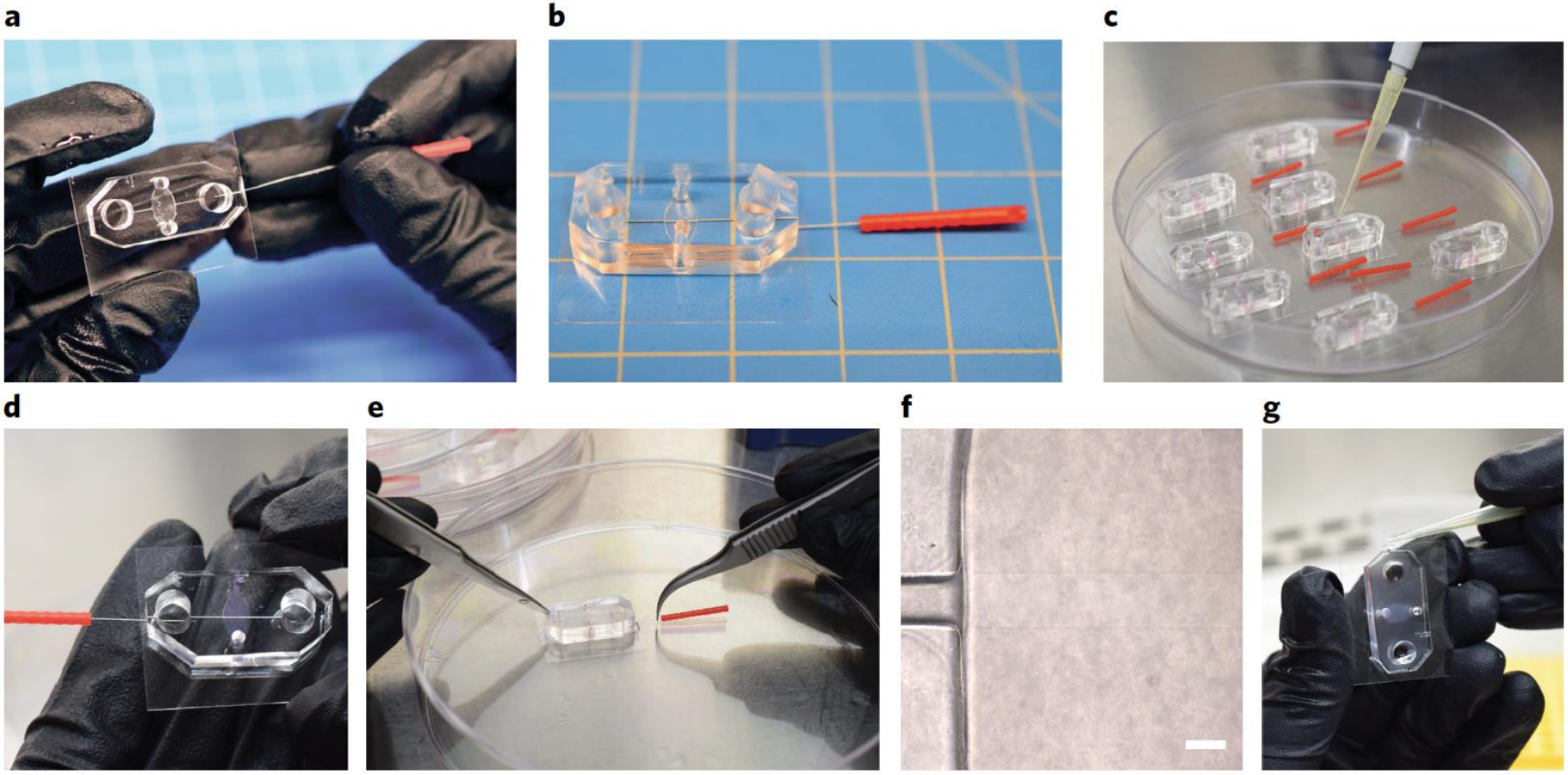

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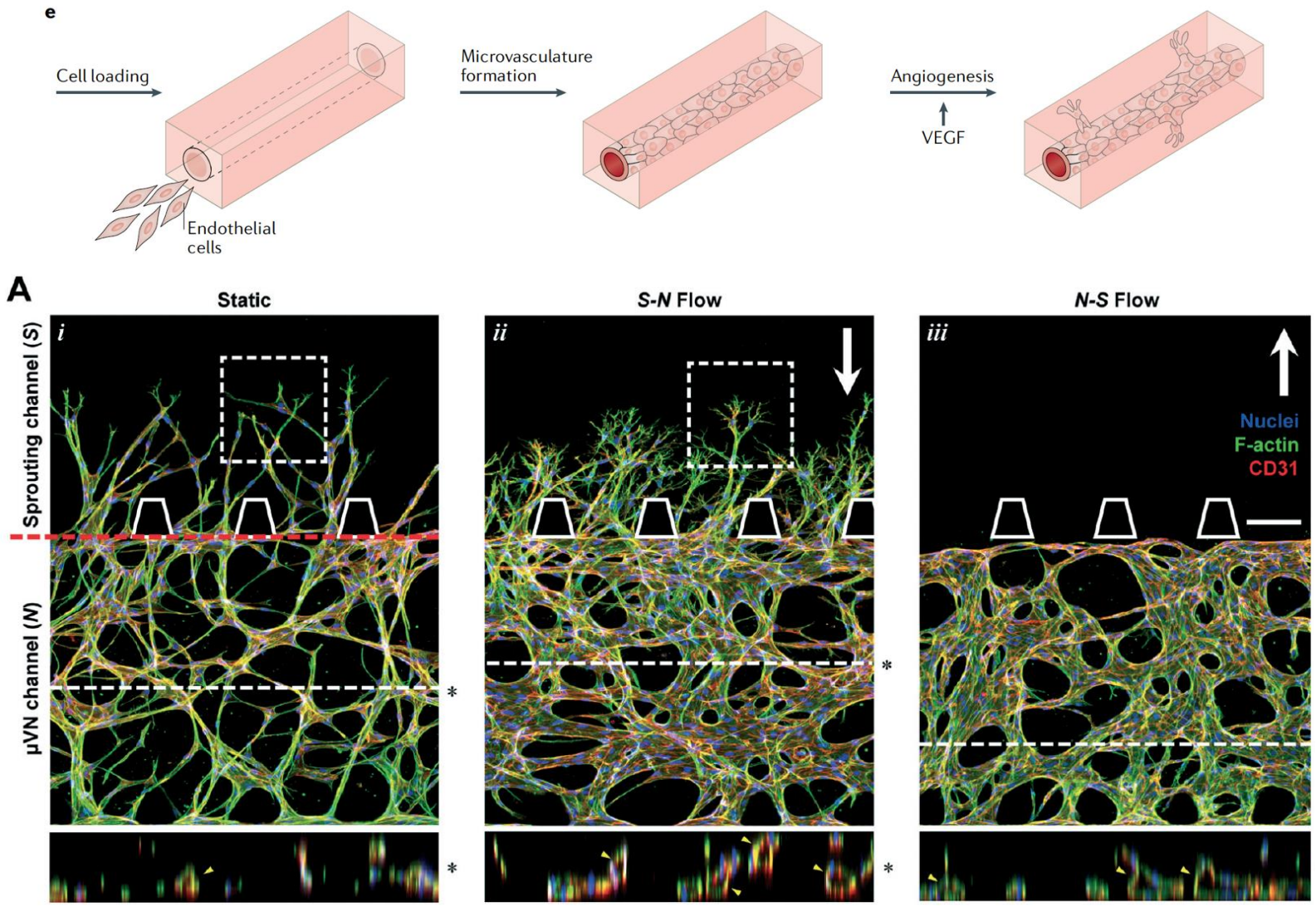
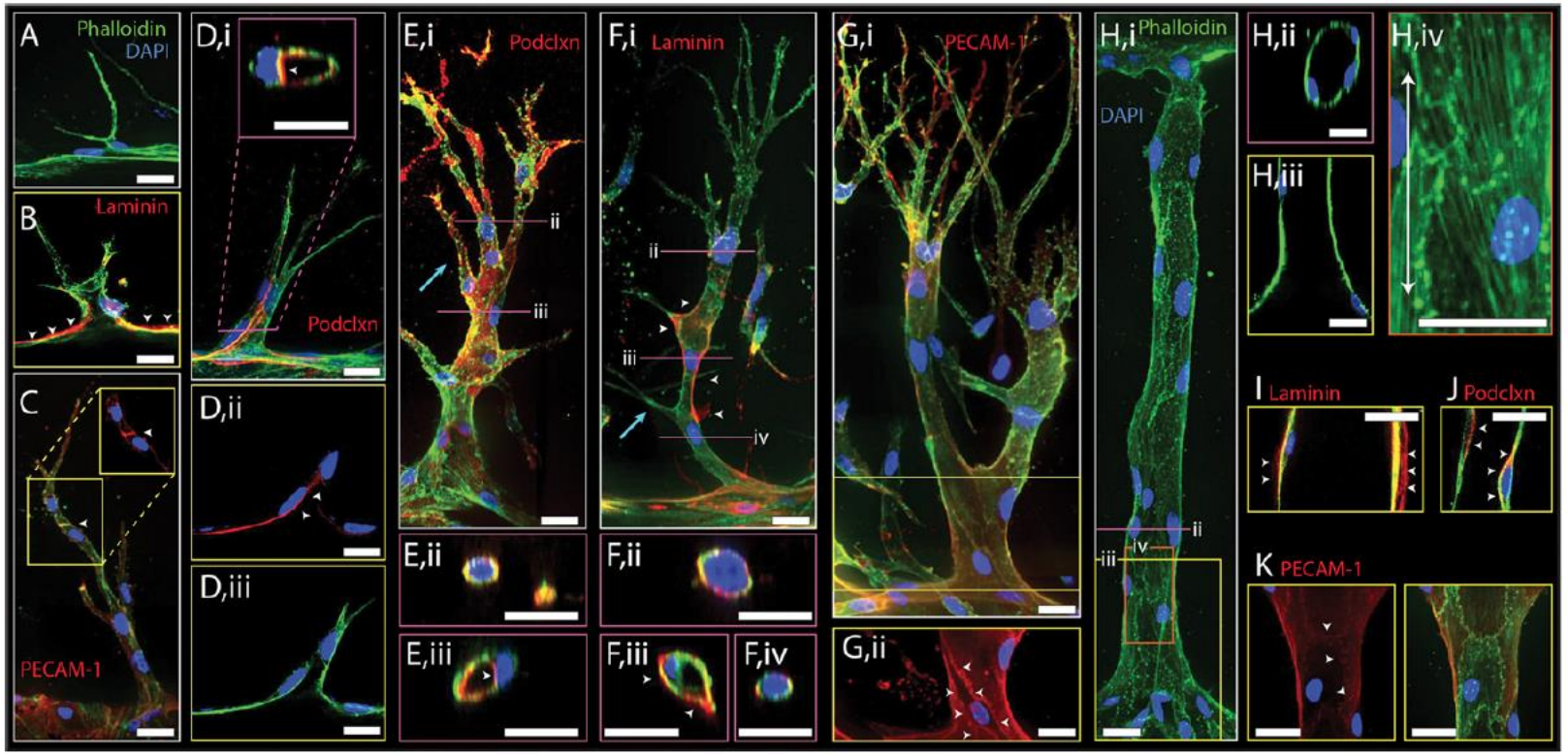


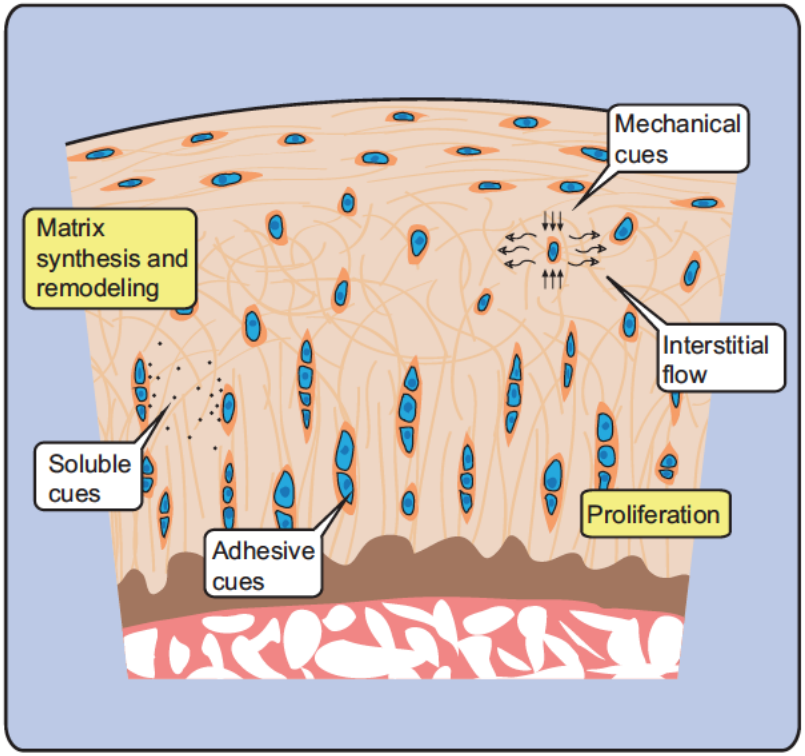
Microfabricated blood vessels

- Angiogenic sprouting
- Nascent vs mature vessels
- Leader cells and follower cells (mesenchymal vs epithelial phenotype)

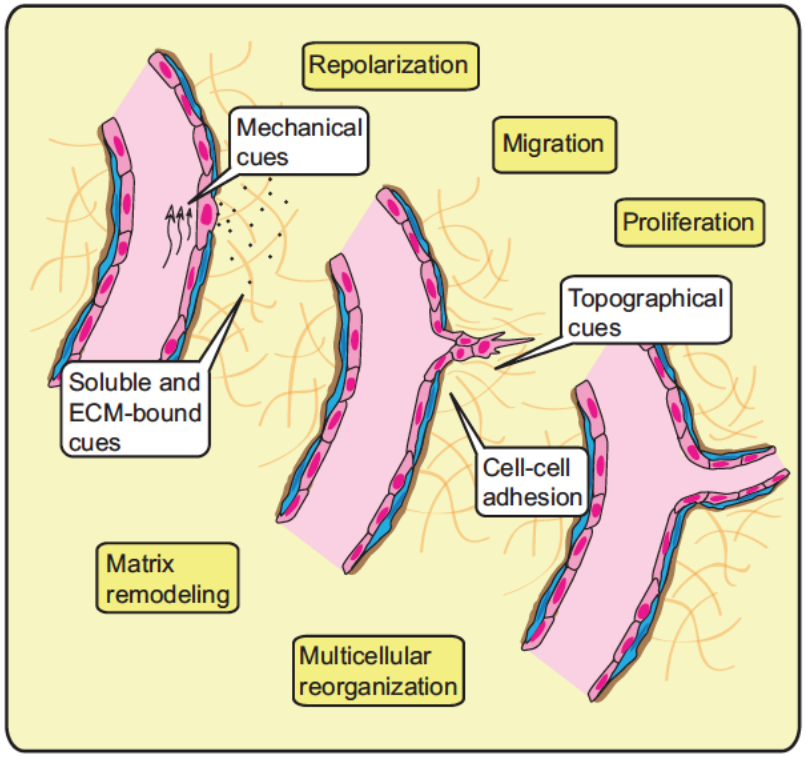


Matrix Mechanobiology

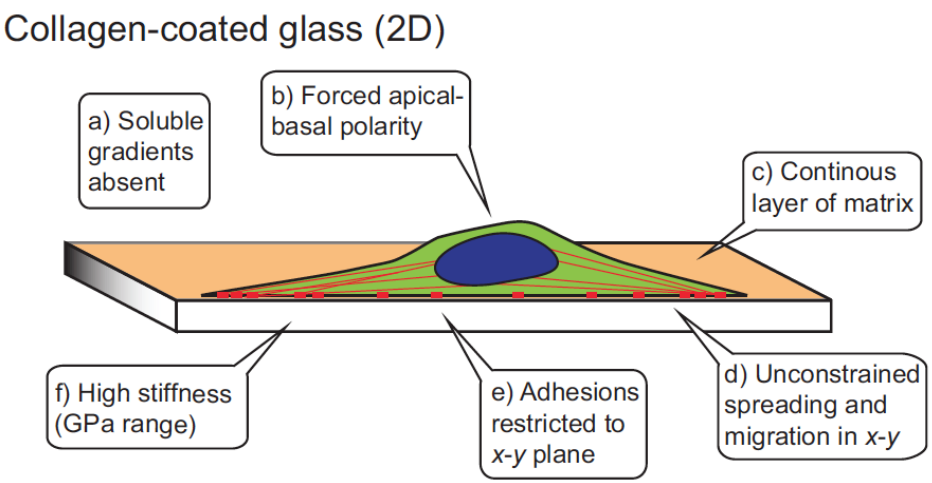
Cartilage and chondrogenesis

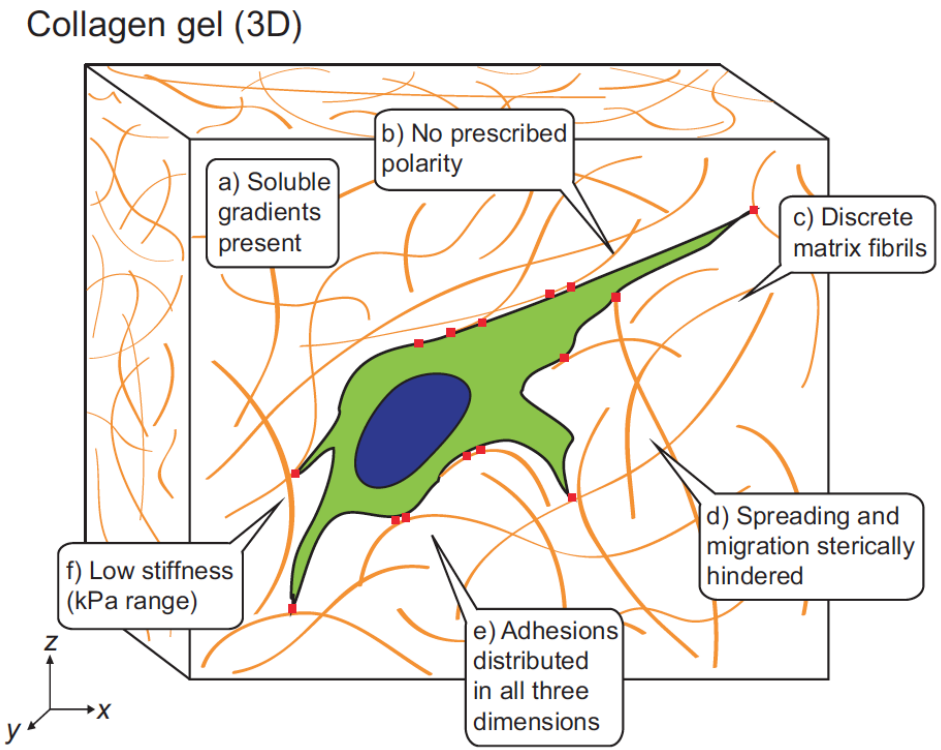


Angiogenesis

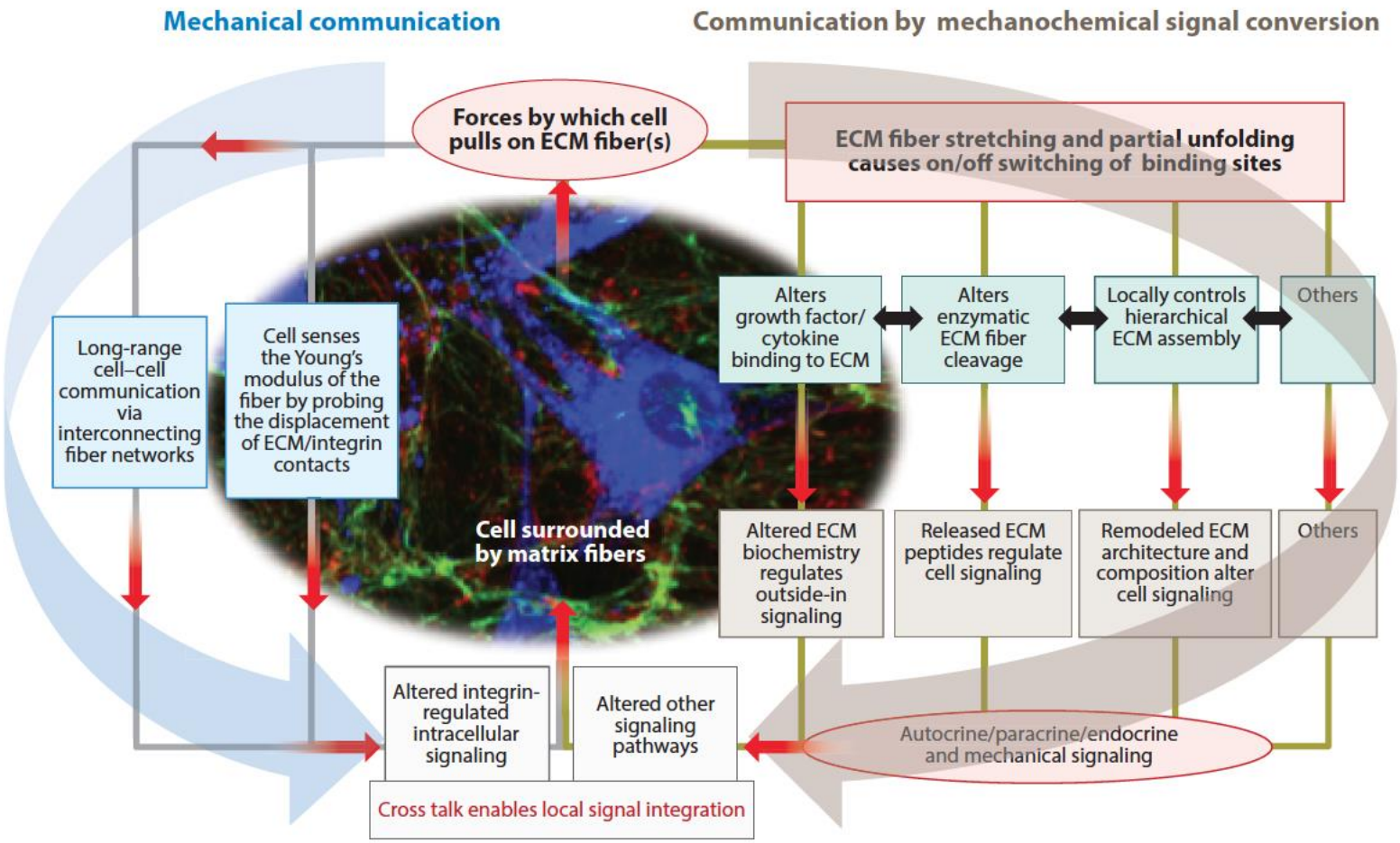


Matrix Mechanobiology





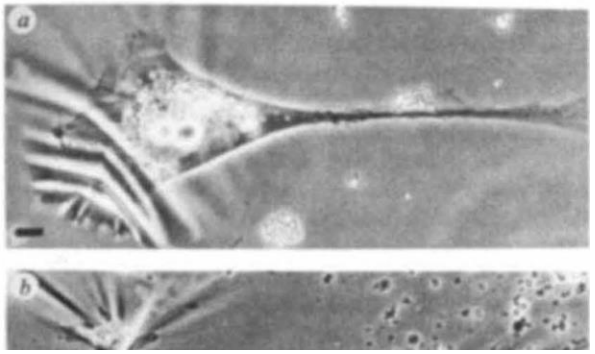
Matrix Mechanobiology



Fibroblast traction and tissue remodelling

findings in S49 cells suggest a new and somewhat
approach for the xenogenization of malignant lympho
Here, the isolation of immunogenic, non-tumorigenic
based on a simple selection for substrate adherence. The
of the cell-surface change(s) that took place on transiti
free-floating to substrate-adhering, non-tumorigenic, ir
genic S49 cells is being investigated. Furthermore, it is

e could still distort the rubber sheet, but only when



Culturing cells on thin distortable sheets of silicone rubber

1mm

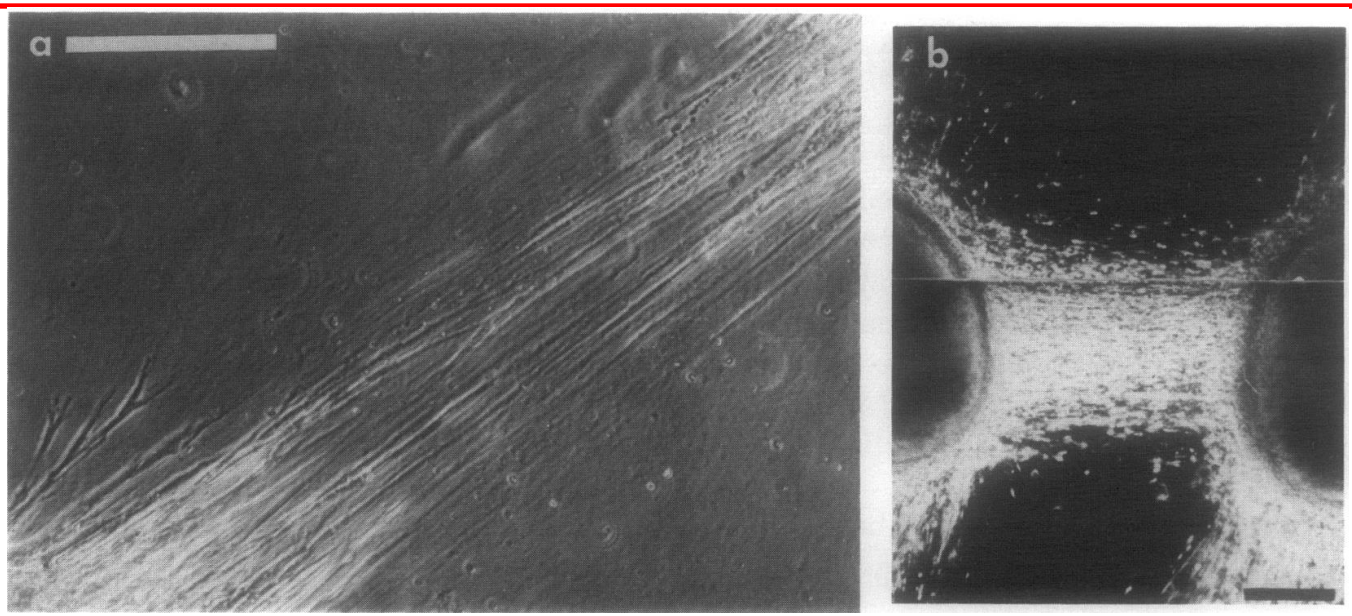
These traction fields resemble Paul Weiss' centre effects, which lends support to his theories of nerve guidance[8] but suggests that guidance should be towards foci of strong traction rather than high growth rate. Our findings also point to fibroblast traction as the cause for the apparent contraction of collagen networks around deep wounds, burns and surgically implanted prostheses[9–11].

We had assumed originally that substratum distortion was merely an incidental by-product of cell locomotion, useful for studying its mechanism. However, these results suggest that traction has evolved to serve morphogenetic functions going

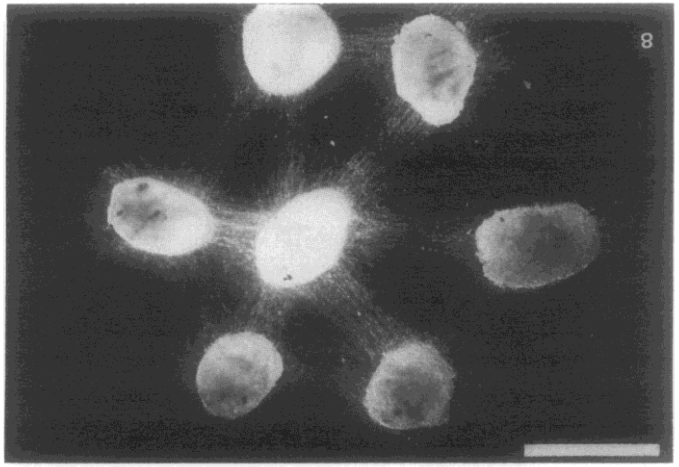
that protective immunity is associated with cell-mediate
mechanisms. We have now examined the capacity of leukocyte
from immune cattle to lyse parasitized lymphoblastoid an
non-parasitized tumour cell lines either directly or after stimu
lation in an autologous mixed lymphocyte reaction (MLR). I
contrast to the nonspecific lytic activity of leukocytes fro
immune cattle reported by Pearson *et al.*[7], we describe th
sequential appearance in the lymph and blood of immune cattl
of cytotoxic leukocytes with activity restricted to target cell
carrying the autologous genotype. These observations sugge
that a major component of protective immunity to *T. parva*

Fibroblasts exert forces very much larger than those actually needed for locomotion. This strong traction distorts collagen gels dramatically, creating patterns similar to tendons and organ capsules. We propose that this morphogenetic rearrangement of extracellular matrices is the primary function of fibroblast traction and explains its excessive strength.

Fibroblast traction and tissue remodelling



The alignment of collagen between explants results in the orientation of fibroblasts migrating from the explants in the same direction.



Cell patterning in ECM

gradient (Carter, 1967; Harris, 1973), contact inhibition and simple forward biasing of cell motion (Trinkaus, 1982; Englander & Davies, 1979; Gail & Boone, 1970; Gould, Selwood, Day & Wolpert, 1974).

While all of these mechanisms probably act at some point in development, it is not clear how they conspire to generate organized spatial aggregations of cells. In this paper we will describe a simple mechanism by which patterned aggregates of motile cells can come about. The mechanism we shall describe is speculative in the sense that we have not demonstrated unequivocally that embryonic pat-

Mesenchymal cells within embryos nearly all have a capacity for **autonomous locomotion** which is analogous to that of amoebae, but is perhaps best described as crawling. The individual cells move by **exerting forces upon their surroundings**, which generally consist of a fibrous extracellular matrix and the surfaces of other cells. These tractions require that the moving cell has established **anchor points** to its substratum. The traction forces are exerted by cellular extensions, often flattened, which Abercrombie termed 'leading lamellae' (Harris, 1983). Most cells have several of these protrusions extending in opposing directions. Since the traction exerted by each is directed centripetally, the result is a **tug-of-war** with net cell displacement occurring at the **direction** of the lamellae with the **strongest tension**, and/or the **strongest adhesions** to the substratum.

Cell patterning in ECM

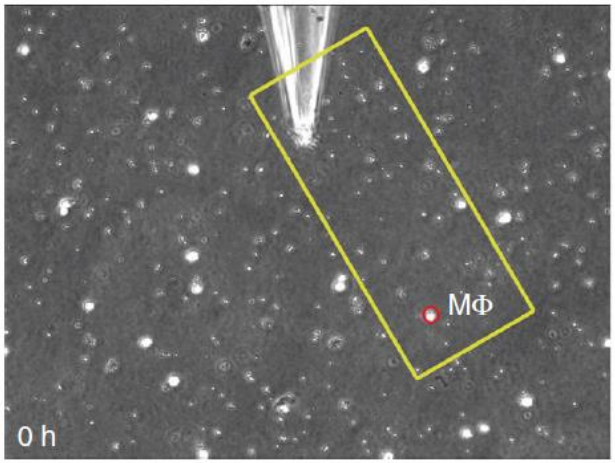
- Cells can be passively dragged along by the contractions of its immediate neighbours, or ride on the substratum which is being dragged by the contractions of distant cells.
- Motile cells will move from less adhesive to more adhesive regions of their substrata
- Numerous experiments have shown that cells will follow geometrical cues in their substratum, such as aligned fibres, grooves, curvature, etc.
- Spreading and migrating cells can exert extremely large traction forces on their substratum.
- A previously isotropic ECM, when seeded with contractile cells, will not remain isotropic: the cells' contractions create strain guidance cues and adhesive gradients which guide the surrounding cells inward toward the centre of contraction.
- The cell tractions are in mechanical equilibrium with the elastic forces of the matrix material. NO INERTIA!
- Within an aligned matrix, such as a collagen gel, cells will configure themselves such that their long axis is in the same direction as the matrix orientation

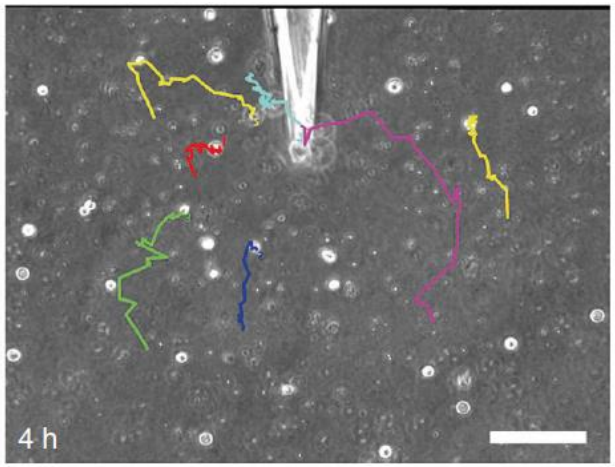
Cell patterning in ECM

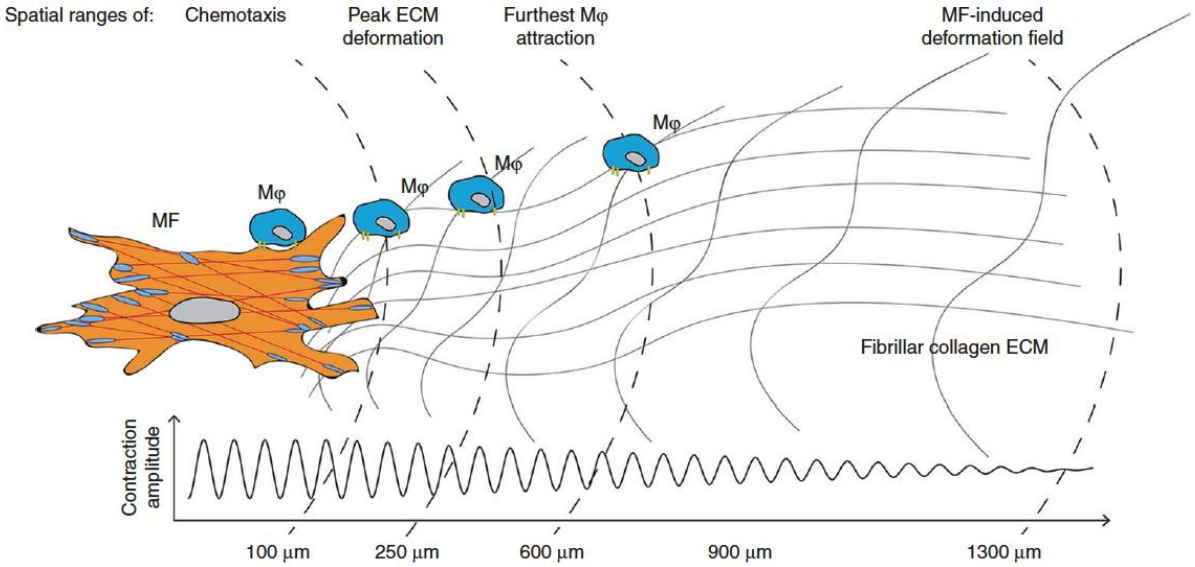
$$\left\{\begin{array}{c}\text{rate of change in matrix density} \\ \text{in a small volume element}\end{array}\right\} = [\text{flux in}] - [\text{flux out}] \qquad (10)$$

ıdding a term to the right-hand side of (10) we could also account for matri
etion by the cells. However, we shall assume for simplicity that, on the time
ɛ of cell movements we are interested in, matrix secretion is negligible.
quations (7), (8) and (10) comprise a complete model for cell motion in ar
tic ECM. While the mathematical structure of the corresponding equations
ved in Appendix A appears complicated (it is), the physical interpretation

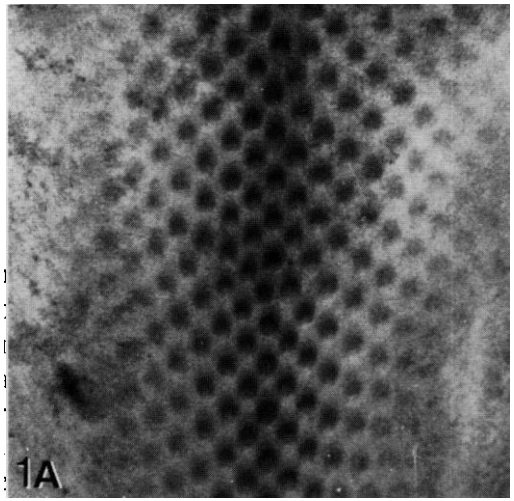
Force-mediated cell recruitment

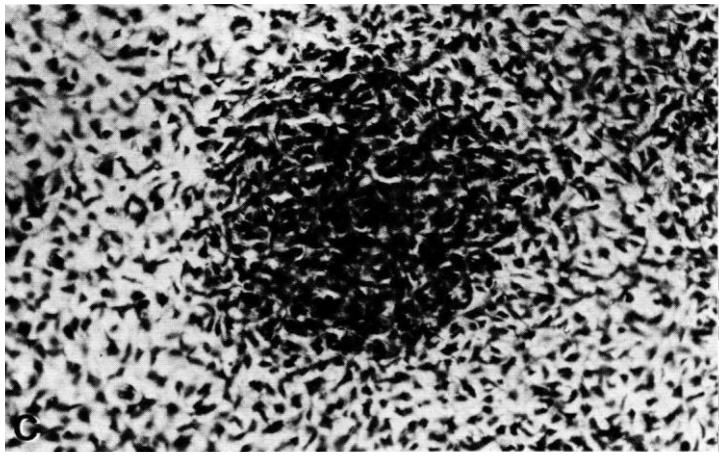


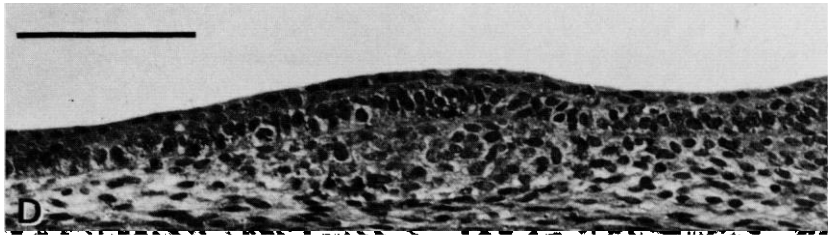


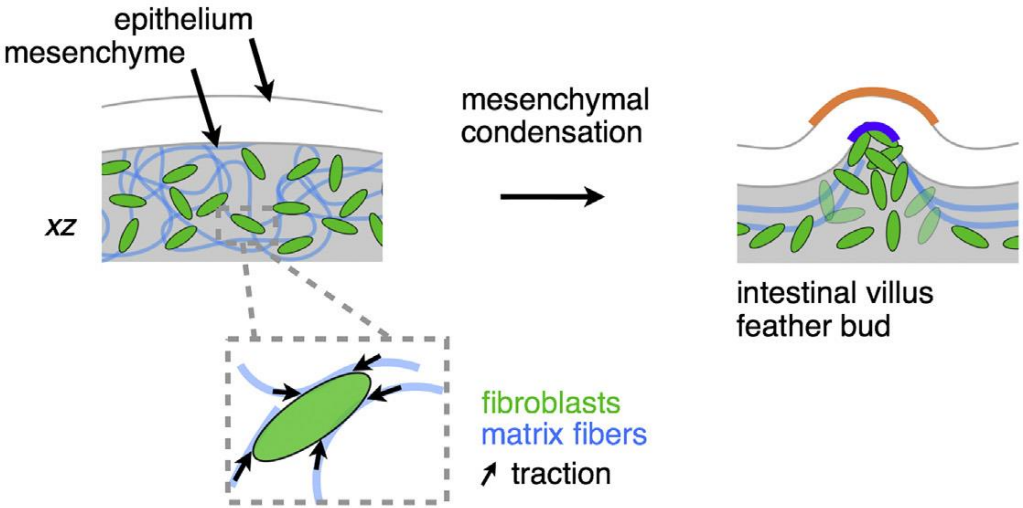


Feather pattern development

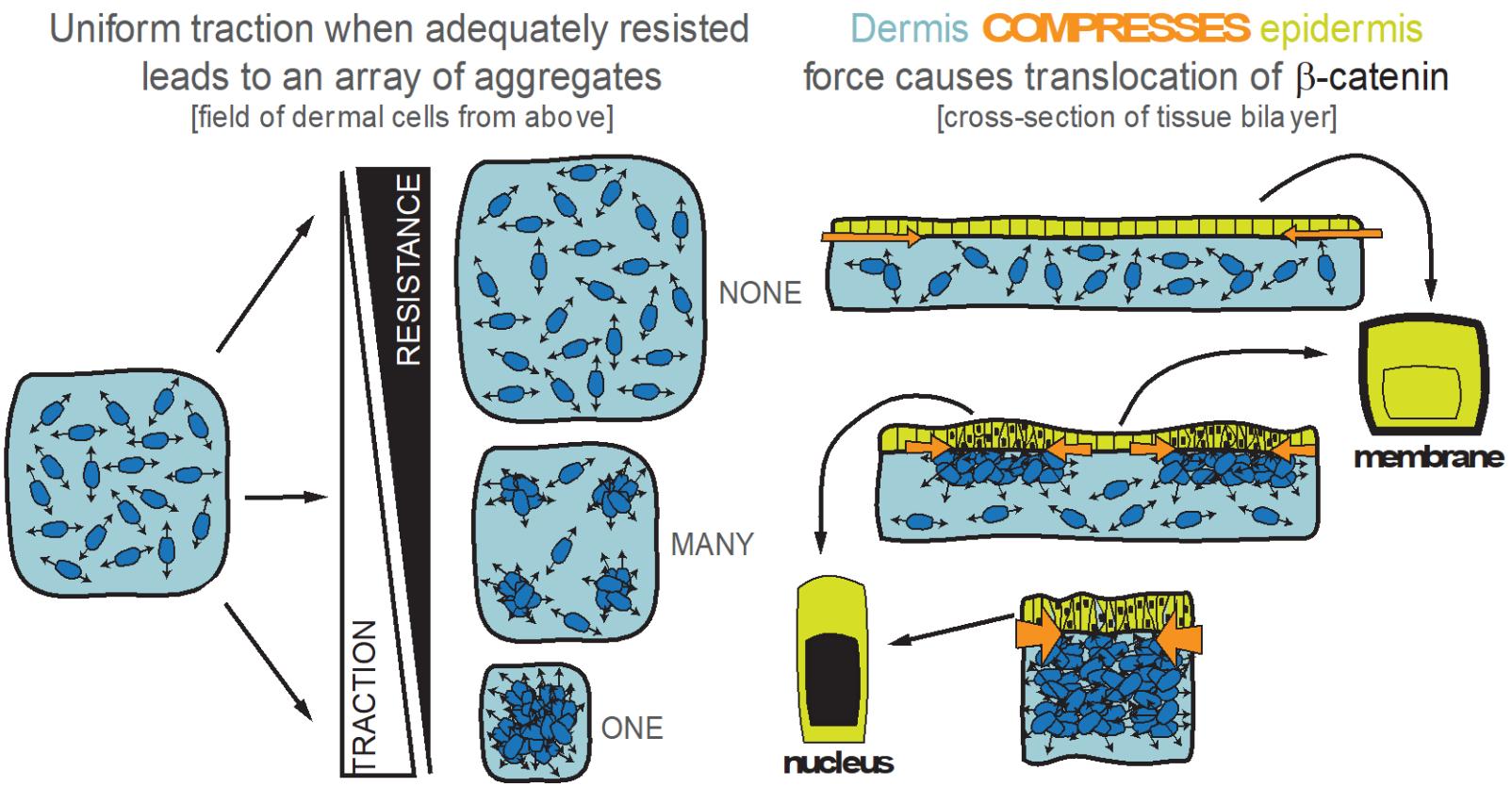








Feather pattern development



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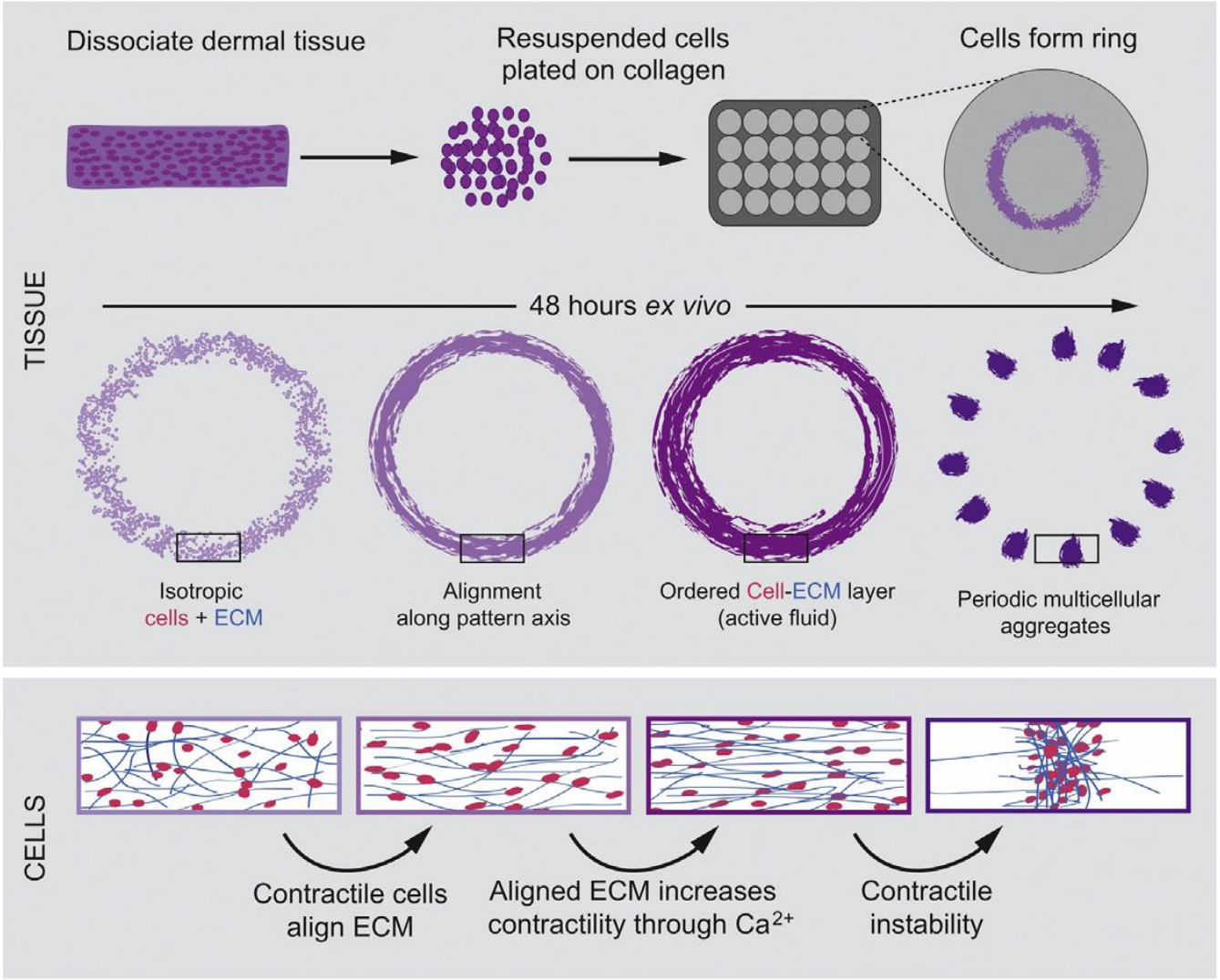
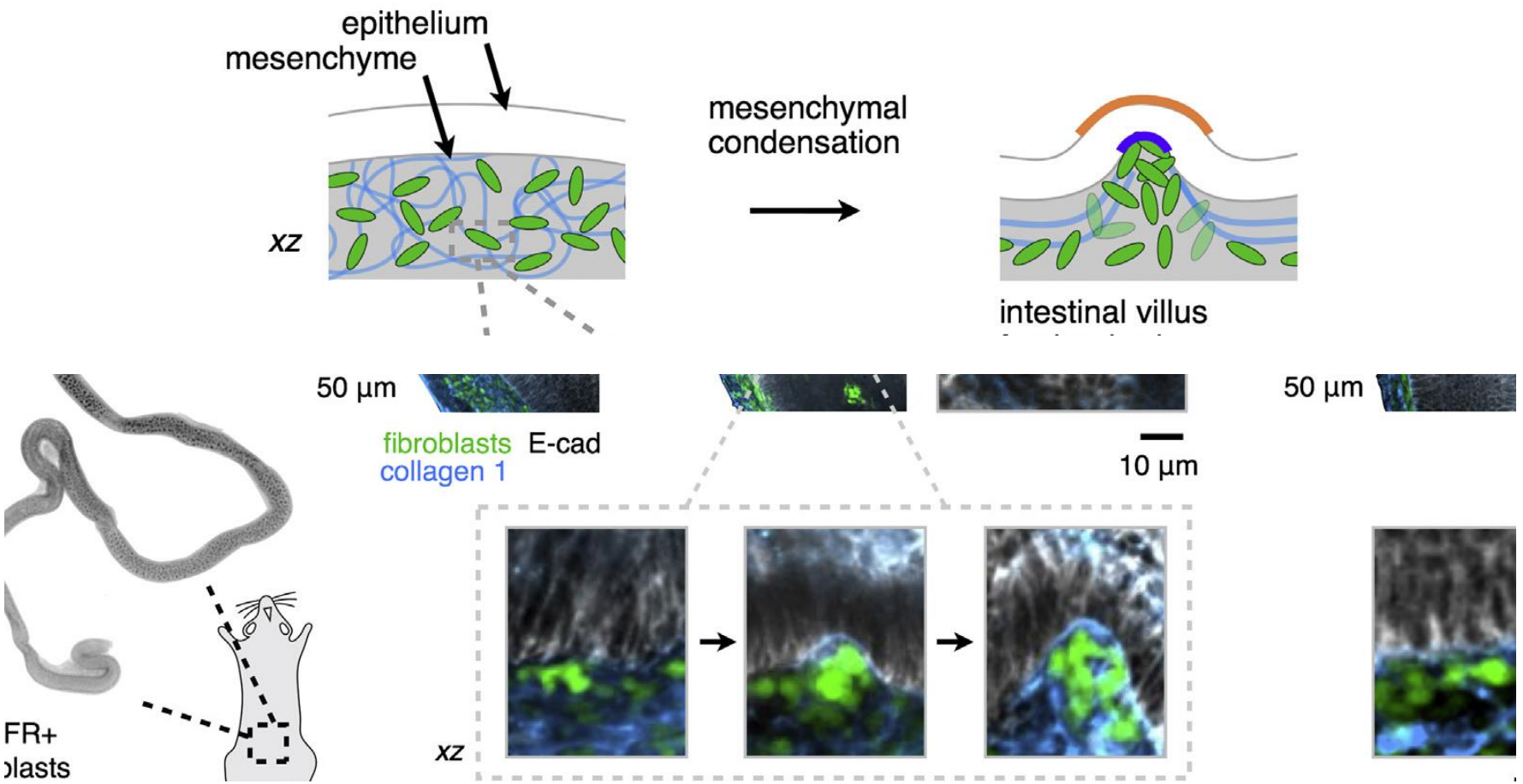
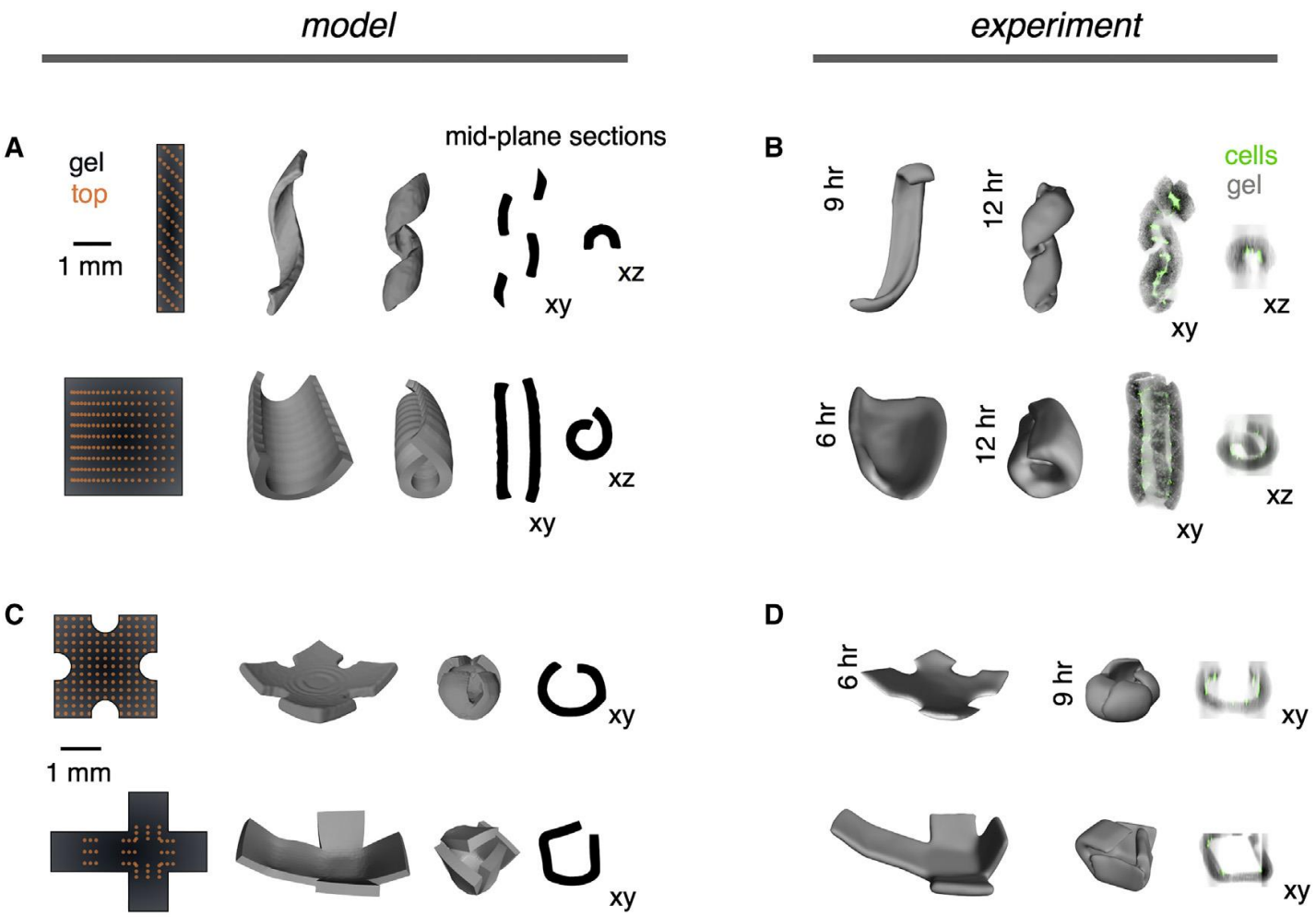


Engineered tissue folding

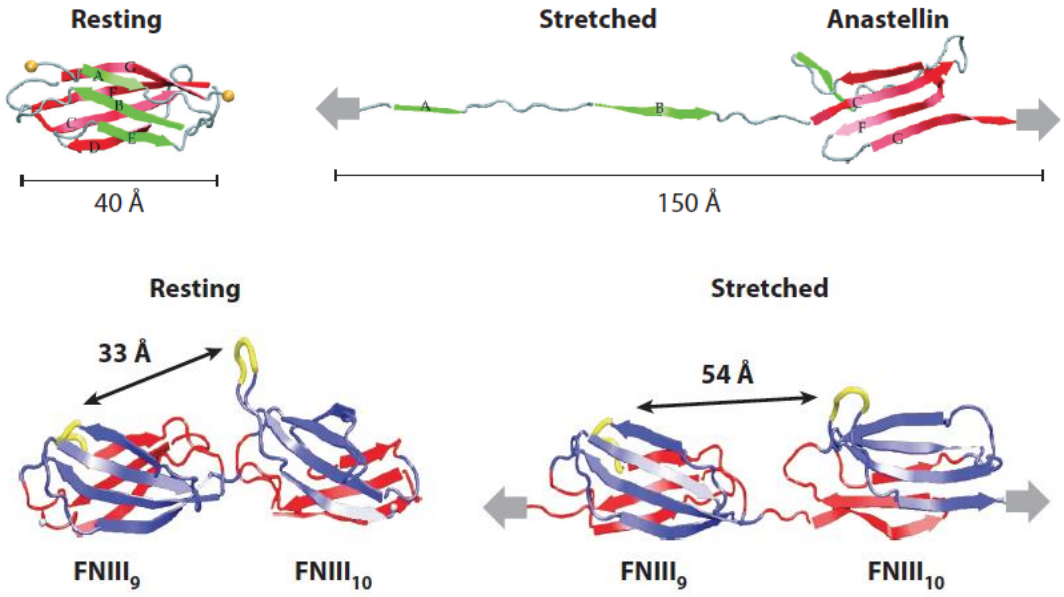


Engineered tissue folding



ECM fibers as mechanochemical switches

- Fiber stretching
  - Increases the Young's modulus of the fiber (strain-stiffening)
  - Activate cryptic sites
  - Activate fibrillogenesis
  - Destroy binding sites
  - Enzymatic degradation



ECM fibers as mechanochemical switches

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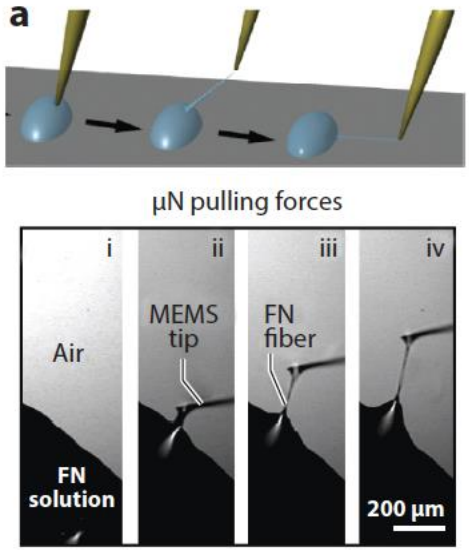
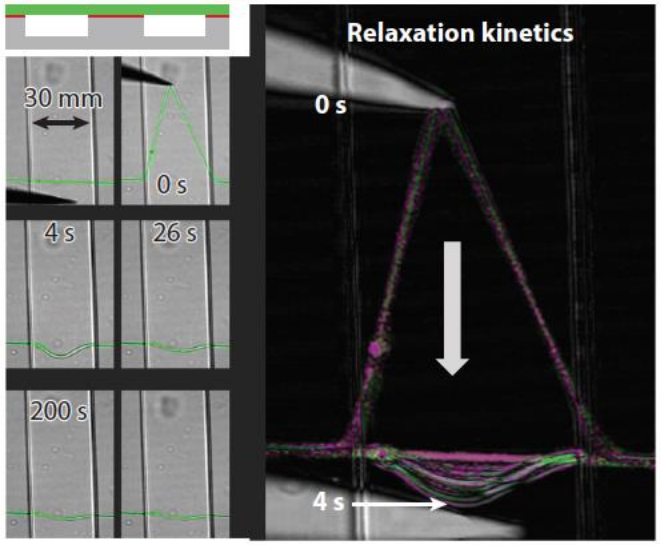
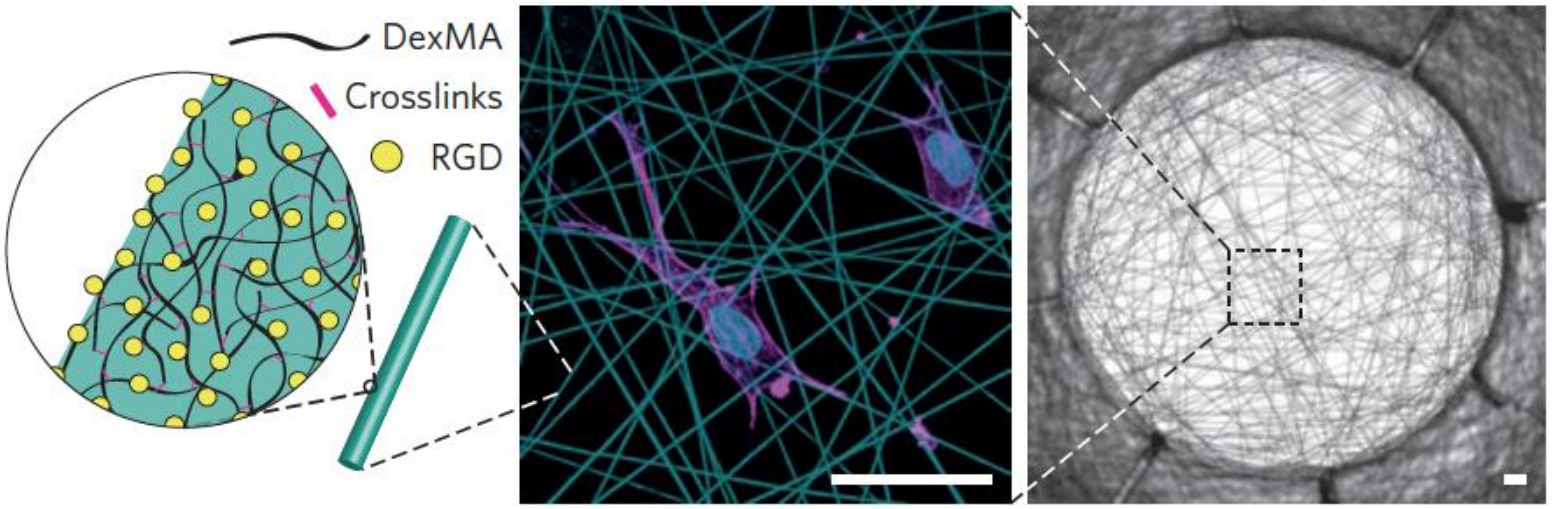




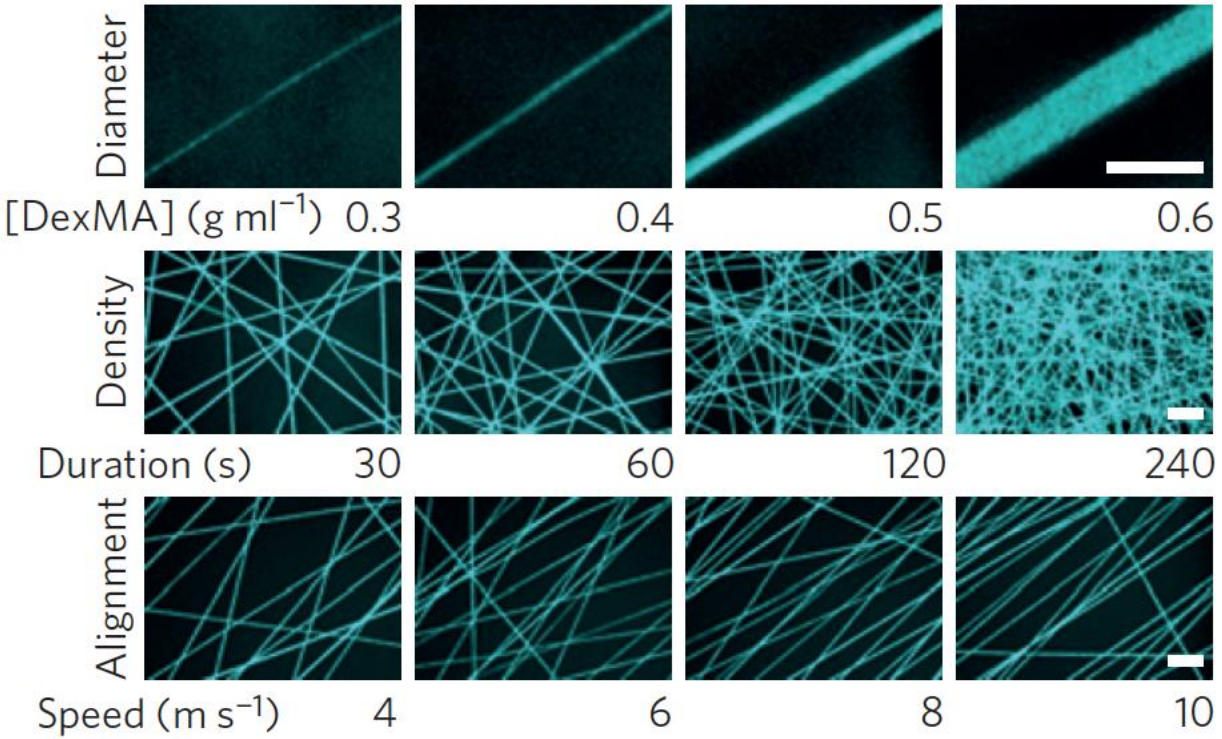
Discrete fiber networks

- Electrospinning Dextran fibers
- Substrates with tunable mechanics and architecture
- Many adjustable parameters: material composition, speed, etc.



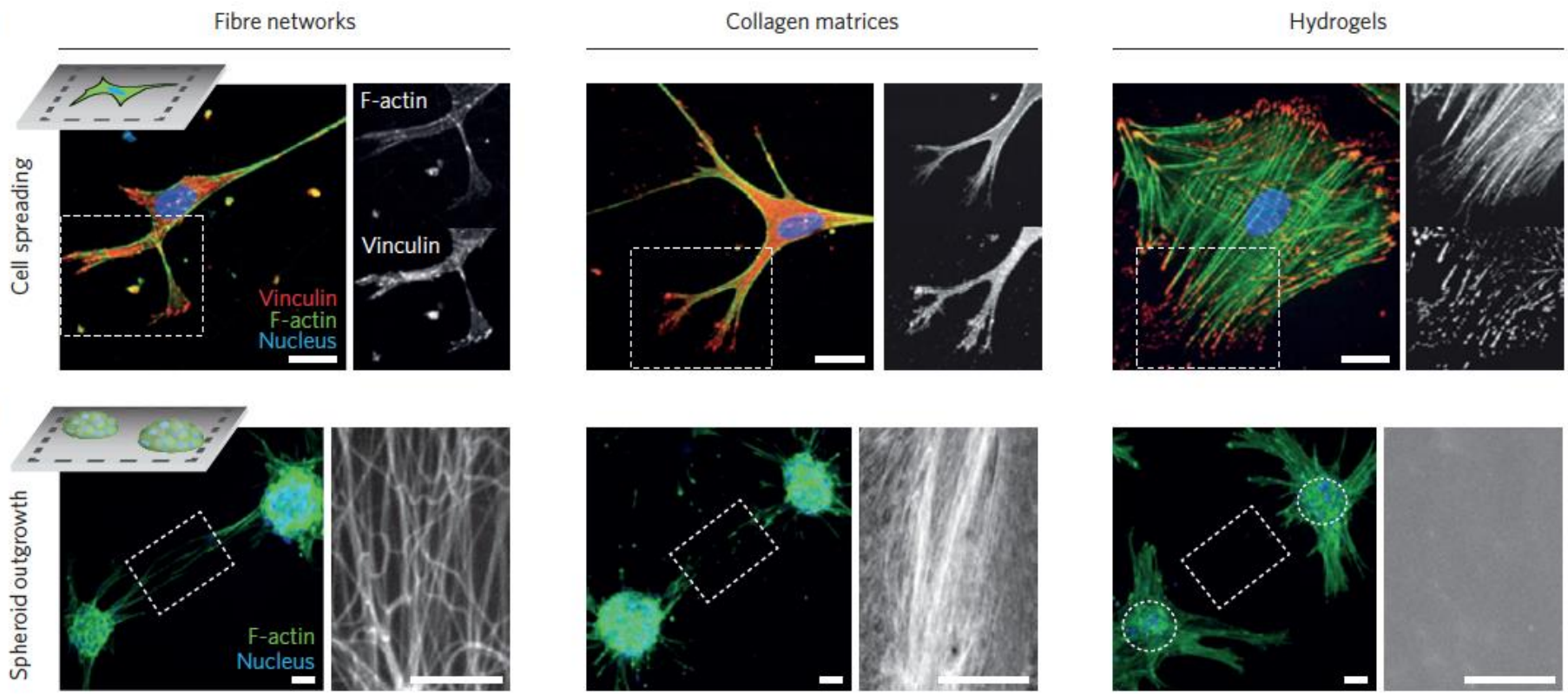
Discrete fiber networks

- Electrospinning Dextran fibers
- Substrates with tunable mechanics and architecture
- Surface functionalization for cell adhesion



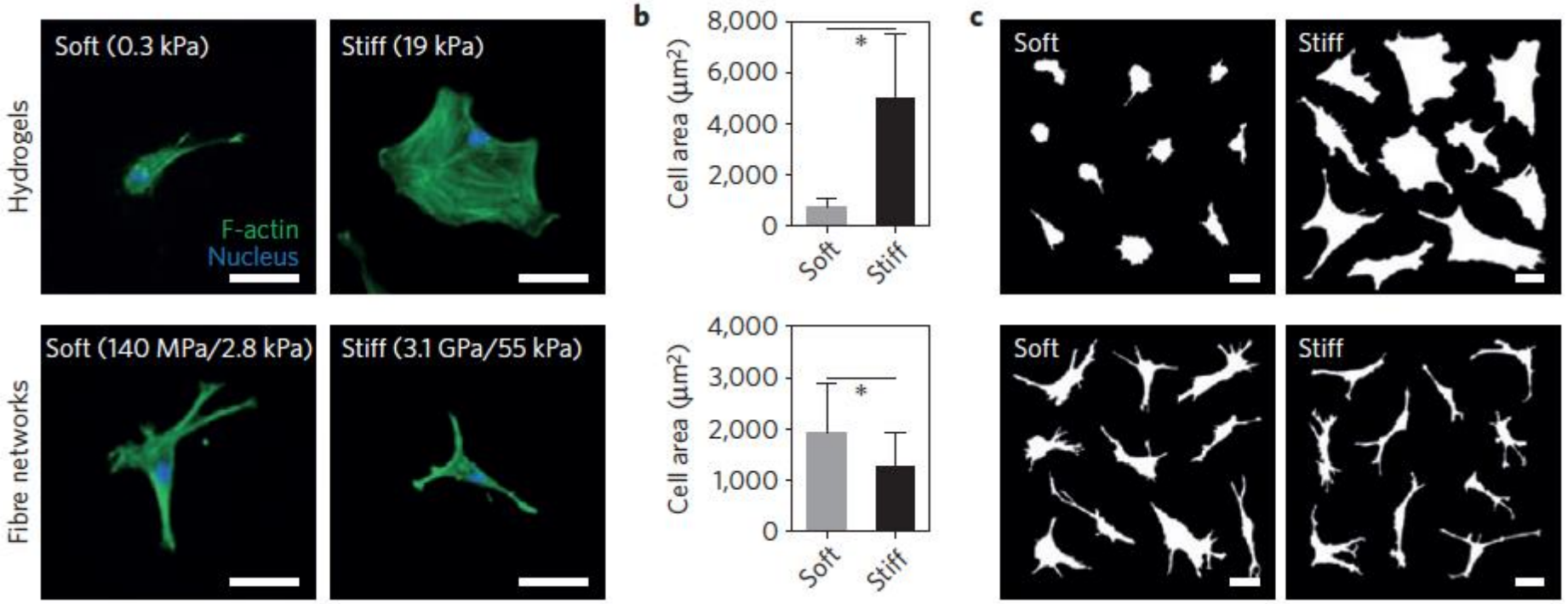
Fiber remodeling

- Single cell and multicellular spreading



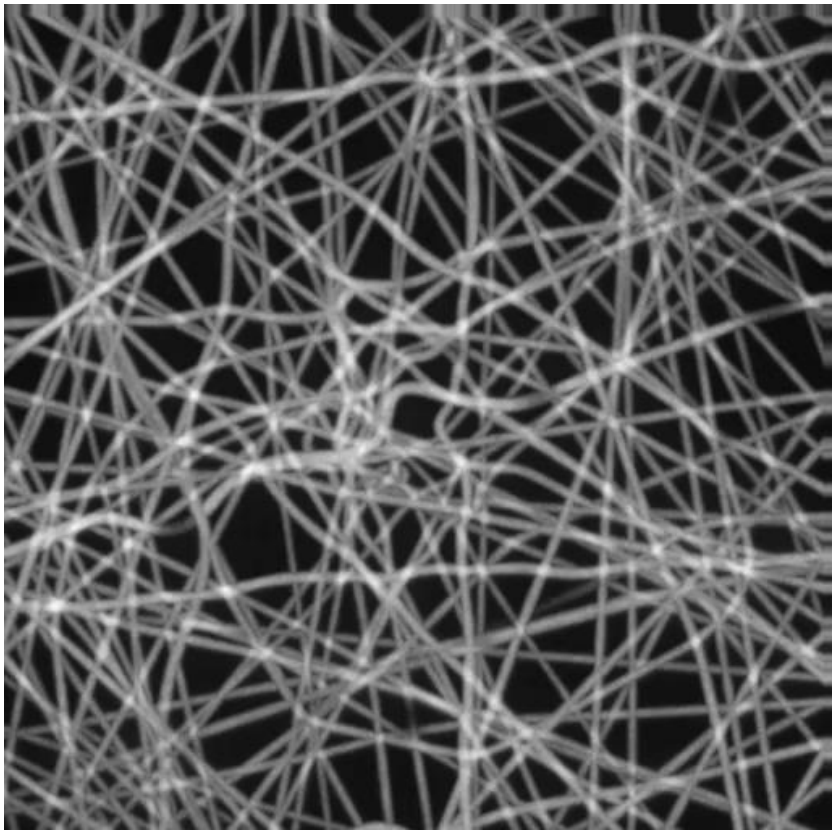
Elasticity

- Soft vs stiff substrates
- 2D hydrogels vs fibrillar matrices



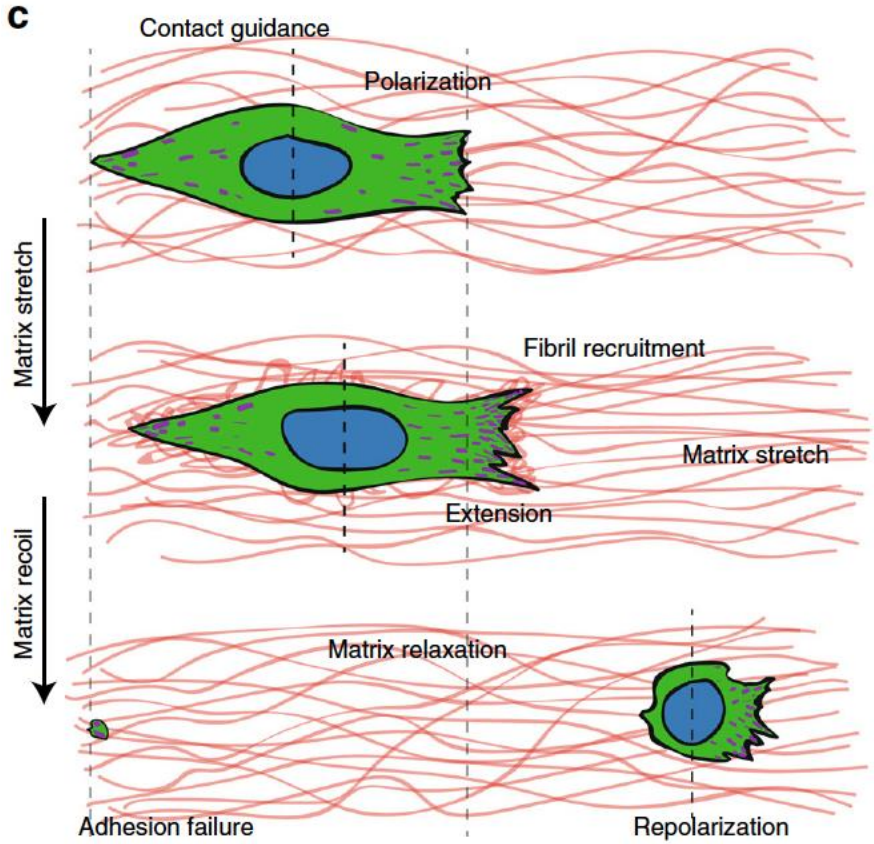
Fiber recruitment

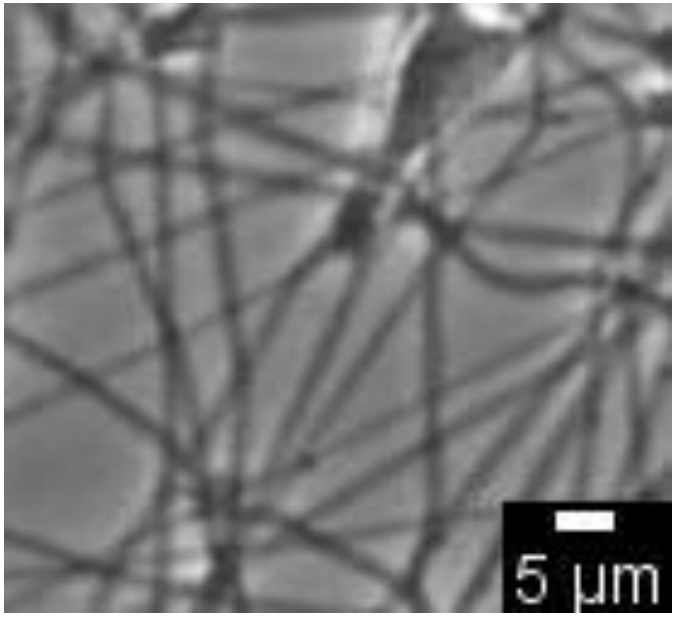
- Pulling fibers, scaffolding, pulling again
- Plastic deformation



Engineered Fibre Networks: Cell migration

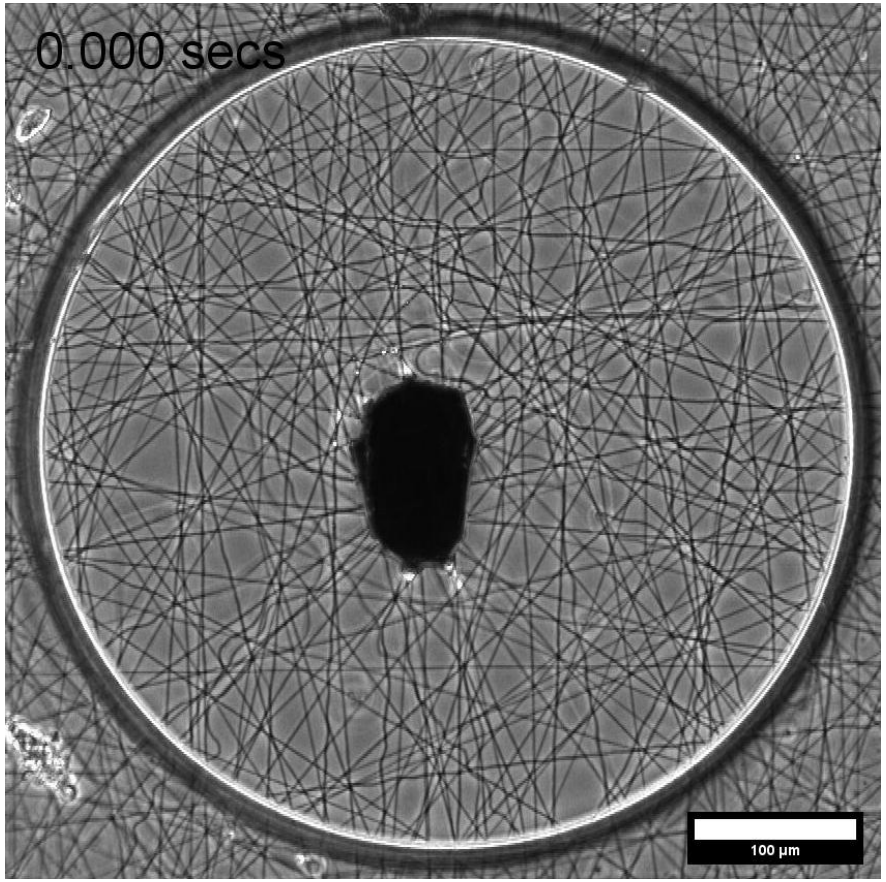
Slingshot migration: elastic energy in migration

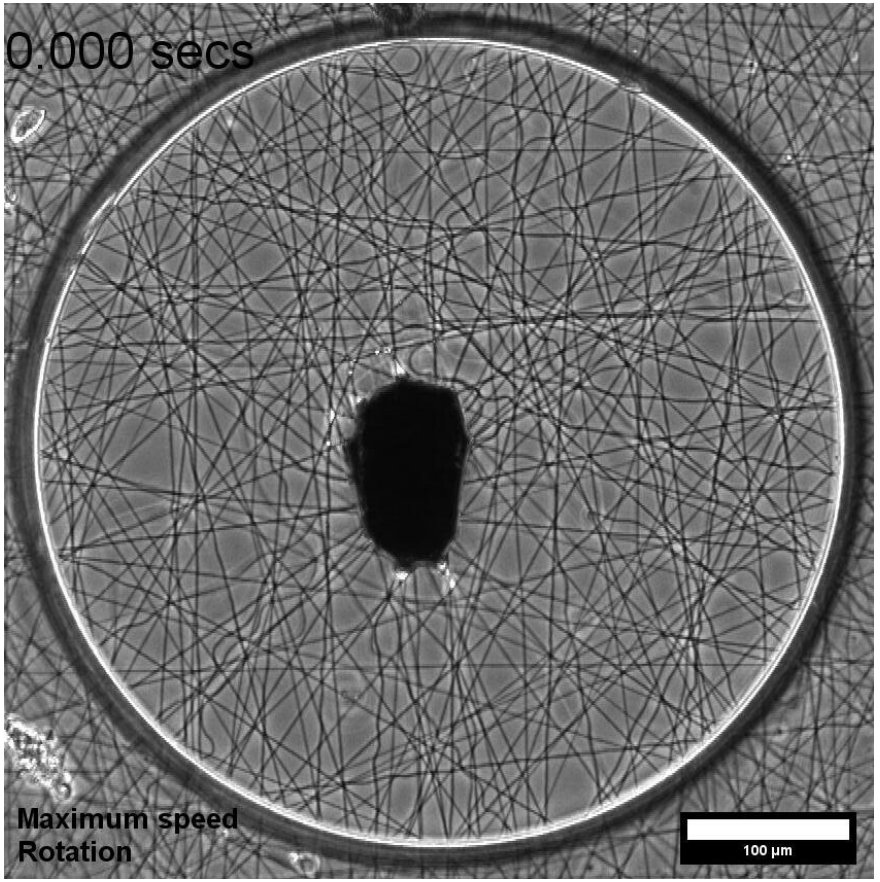




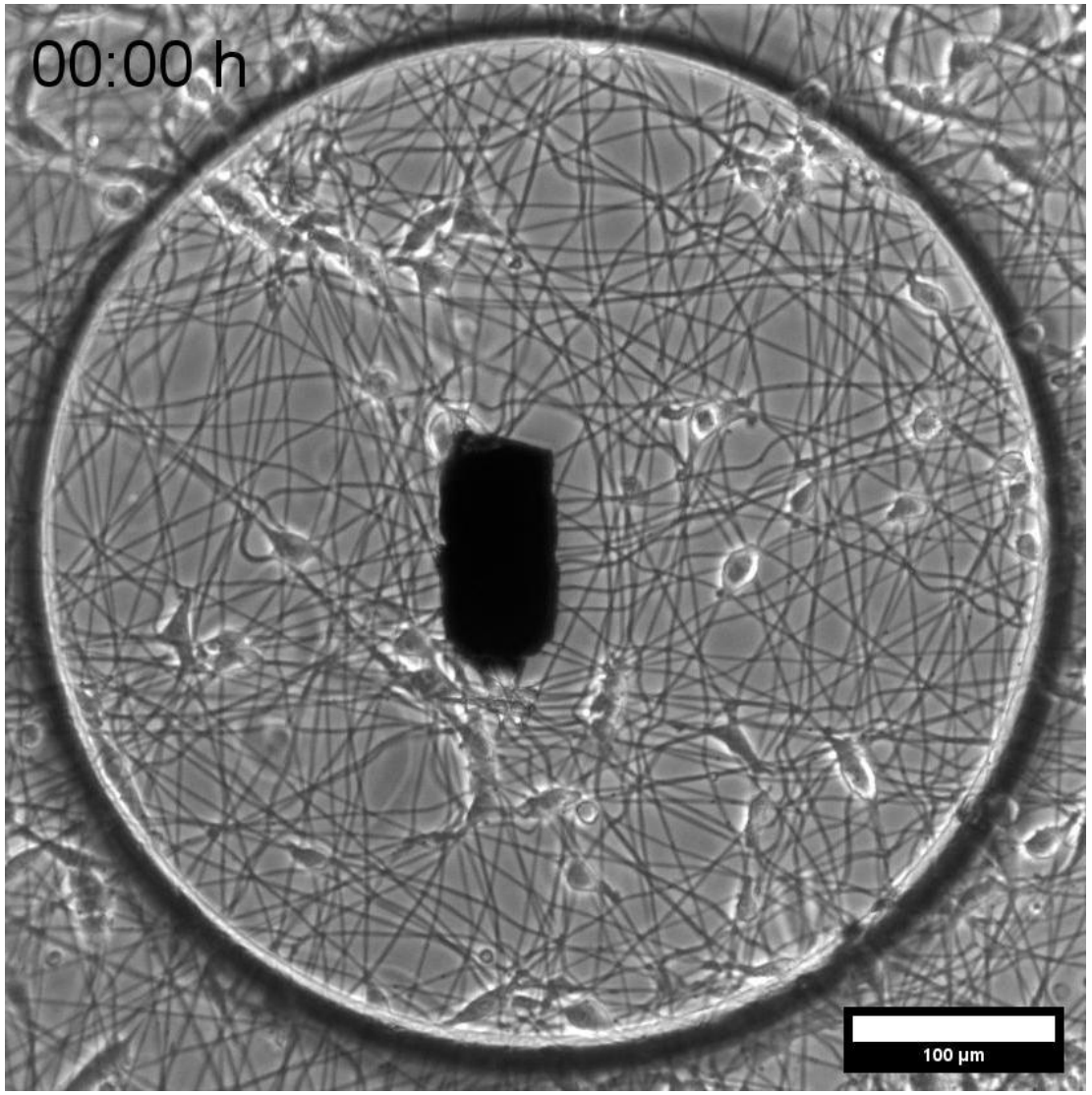
Wang+, **Nat Comm**, 2019

Engineered Fibre Networks: Magnetic manipulation

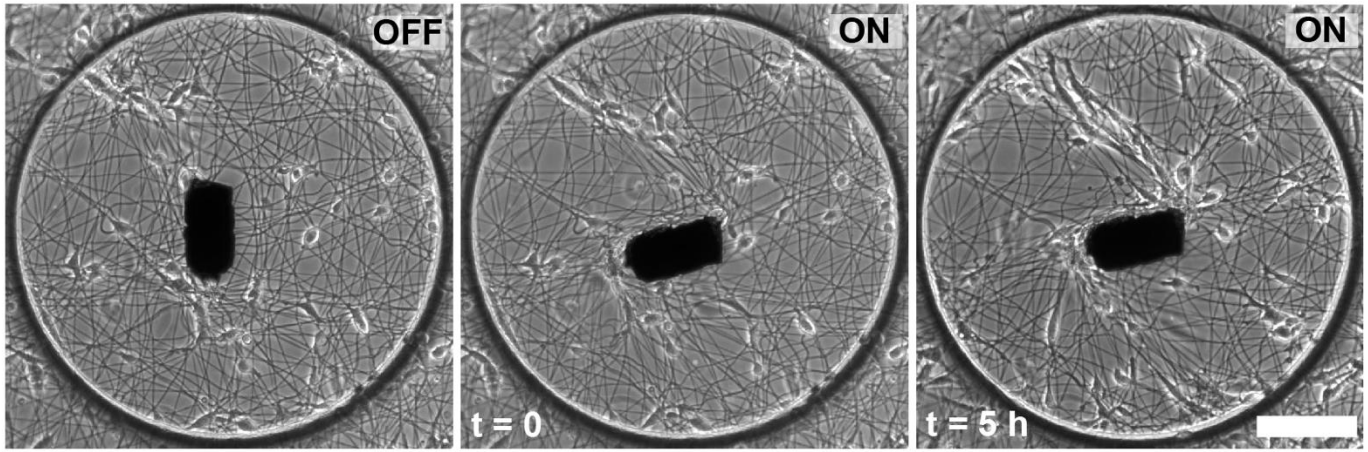


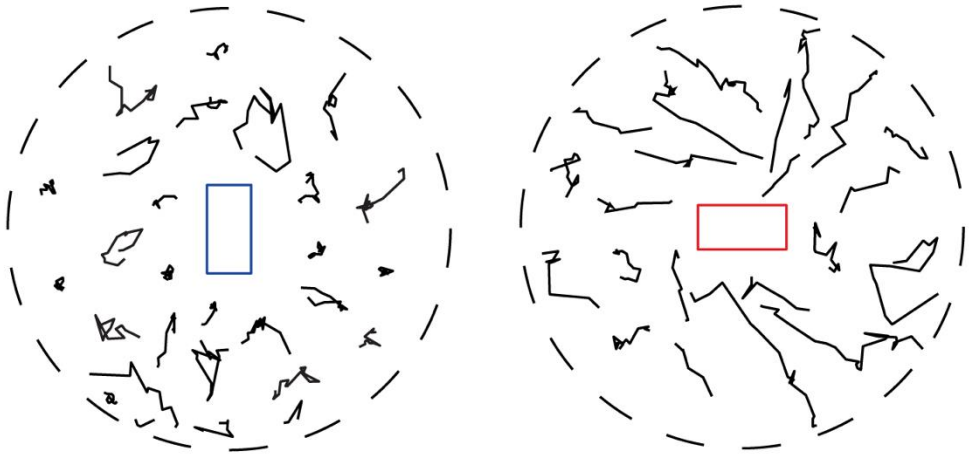


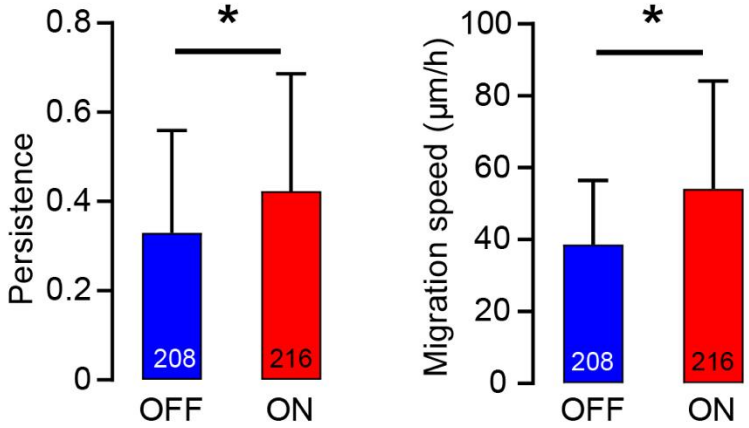
Engineered Fibre Networks: Cell migration



Engineered Fibre Networks: Cell migration

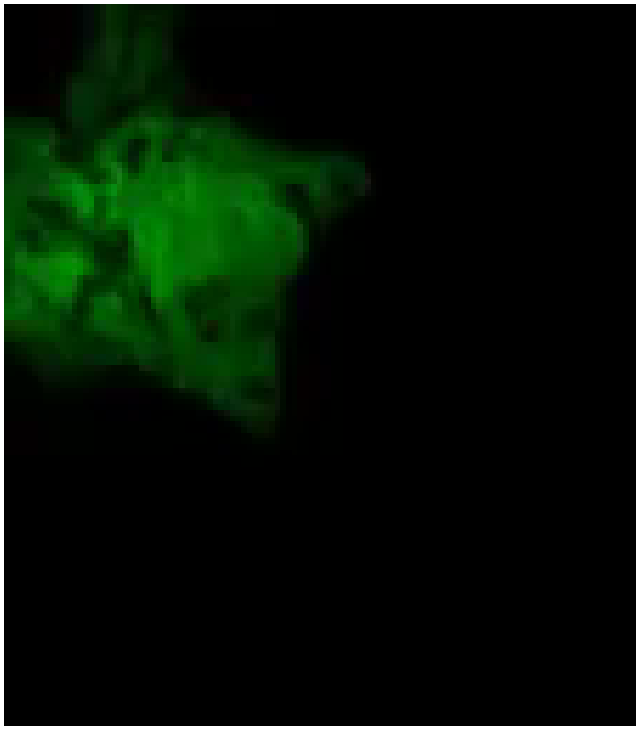


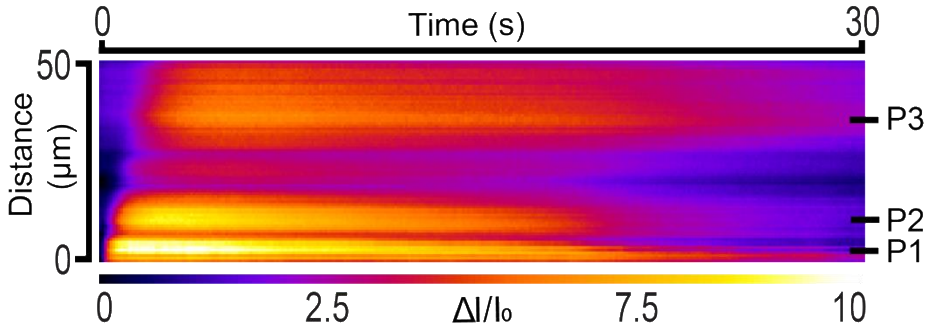


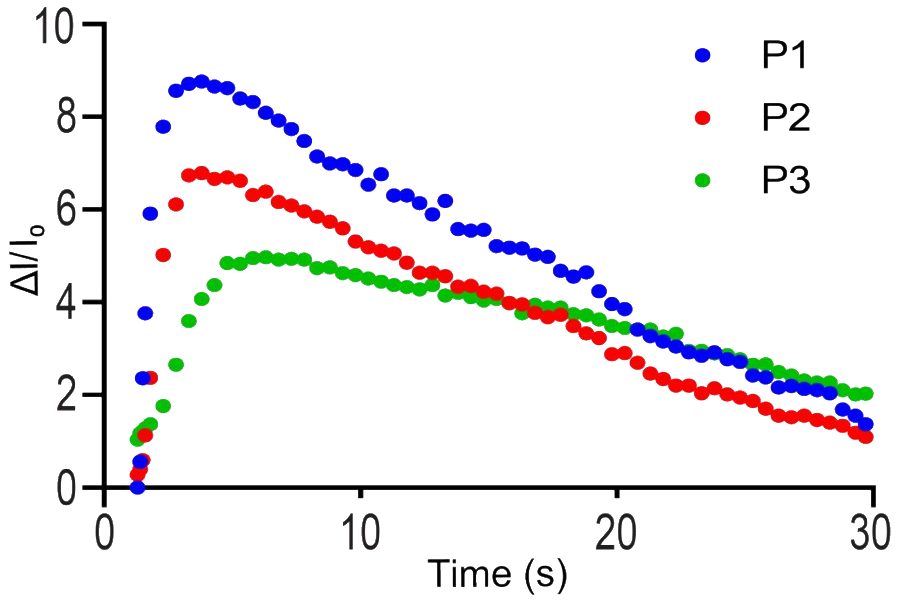


Engineered Fibre Networks: Calcium signaling

Calcium entry and release of stored calcium into the cytoplasm

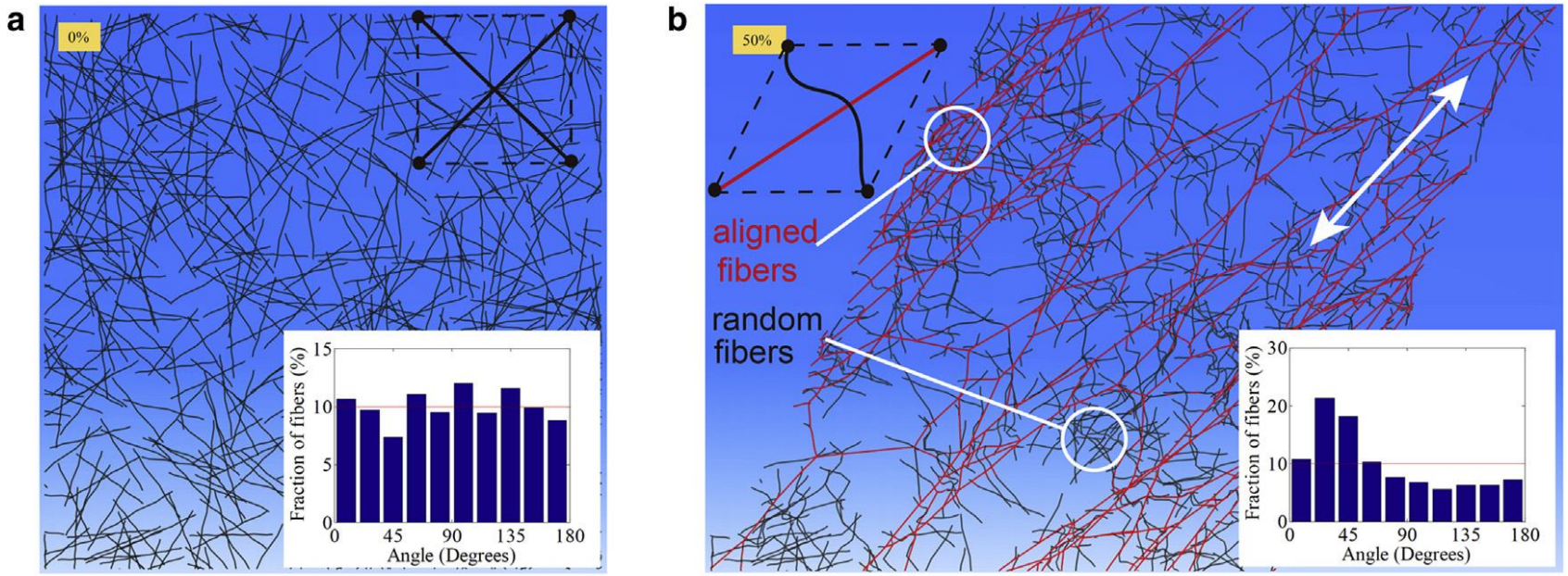






Discrete fibre networks

- Before and after shear deformation



$$W = W_{\mathrm{b}} + W_{\mathrm{f}}$$

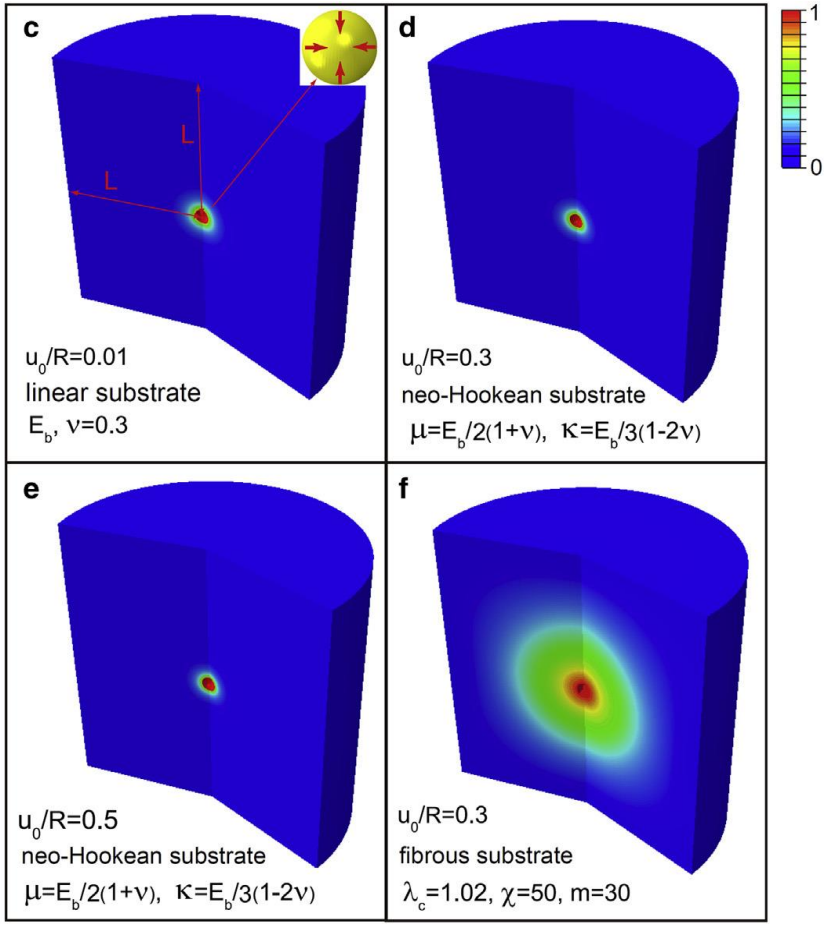
$$W_{\mathrm{b}} = \frac{\mu}{2}\left(\bar{I}_1 - 3\right) + \frac{\kappa}{2}(J-1)^2$$

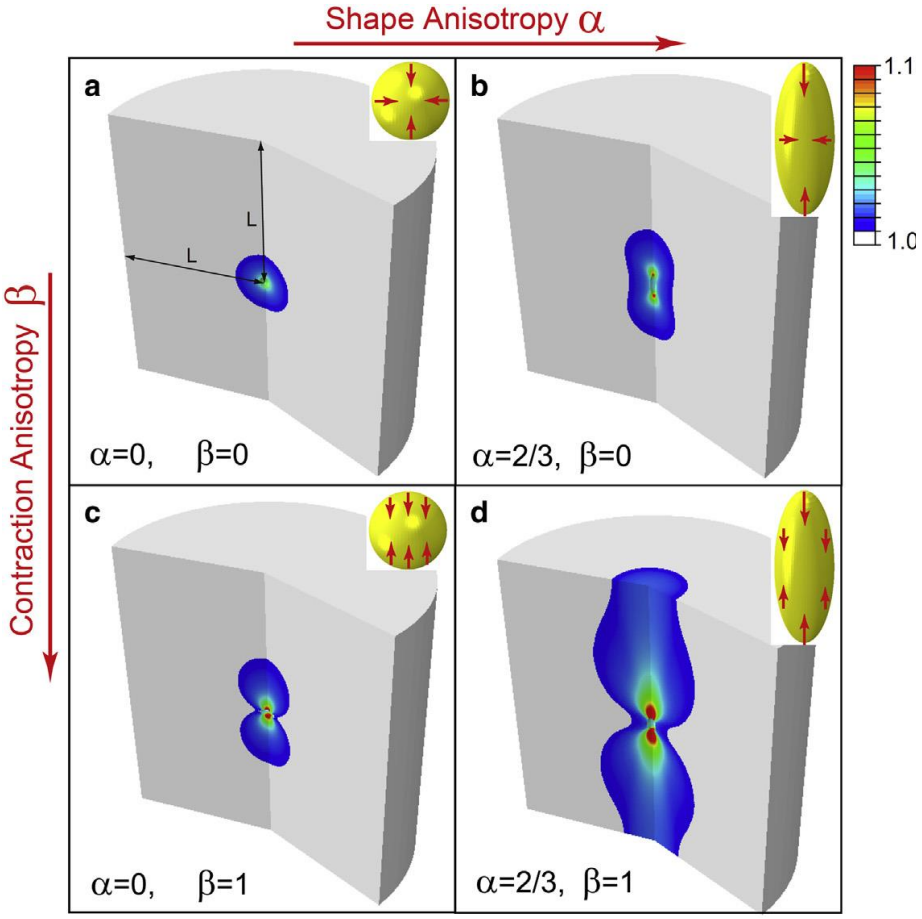
$$W_{\mathrm{f}} = \sum_{a=1}^{3} f(\lambda_a).$$

$$\frac{\partial f(\lambda_a)}{\partial \lambda_a} = \begin{cases} 0 & \lambda_a < \lambda_1 \\ \dfrac{E_f\left(\dfrac{\lambda_a - \lambda_1}{\lambda_2 - \lambda_1}\right)^n (\lambda_a - \lambda_1)}{n+1}, & \lambda_1 \le \lambda_a < \lambda_2 \\ E_f\left[\dfrac{\lambda_2 - \lambda_1}{n+1} + \dfrac{(1+\lambda_a-\lambda_2)^{m+1}-1}{m+1}\right], & \lambda_a \ge \lambda_2, \end{cases}$$

Discrete fibre networks

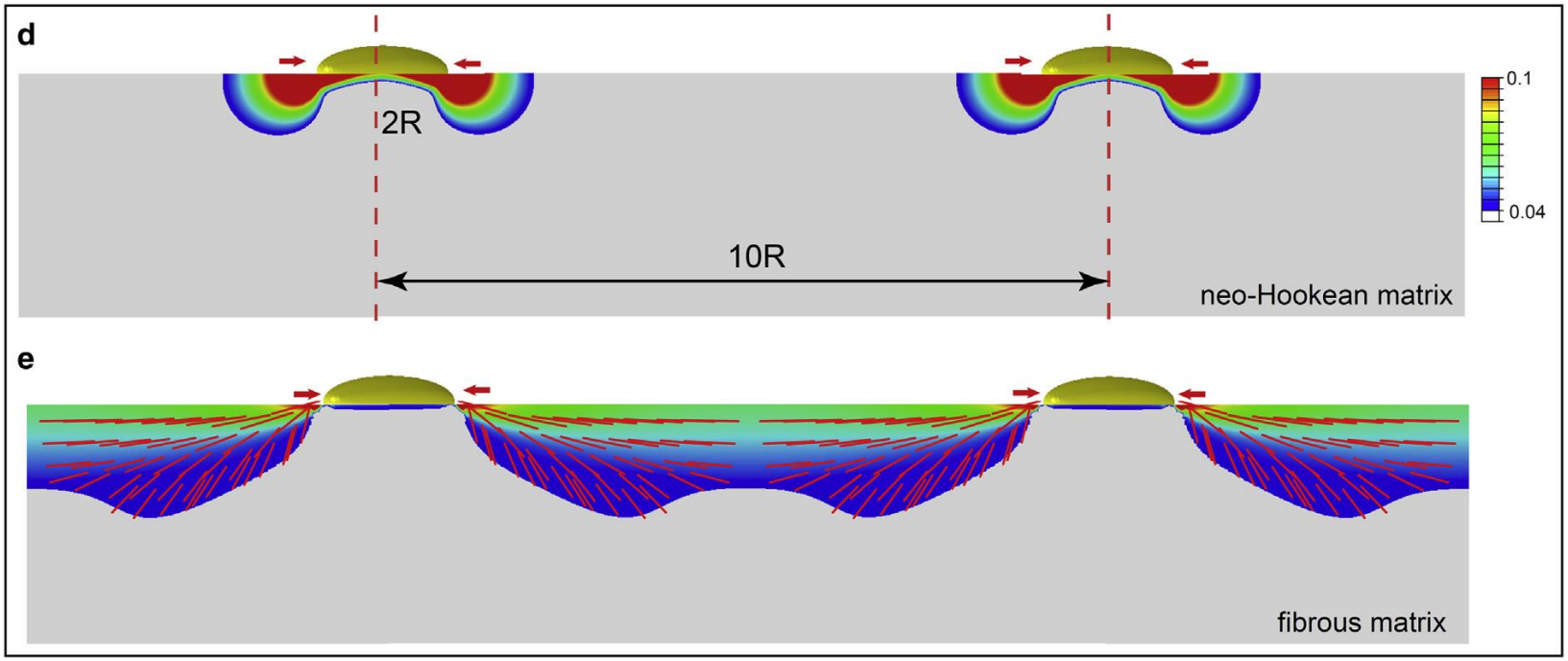
- Color: Maximum principal stretch





Long range force transmission

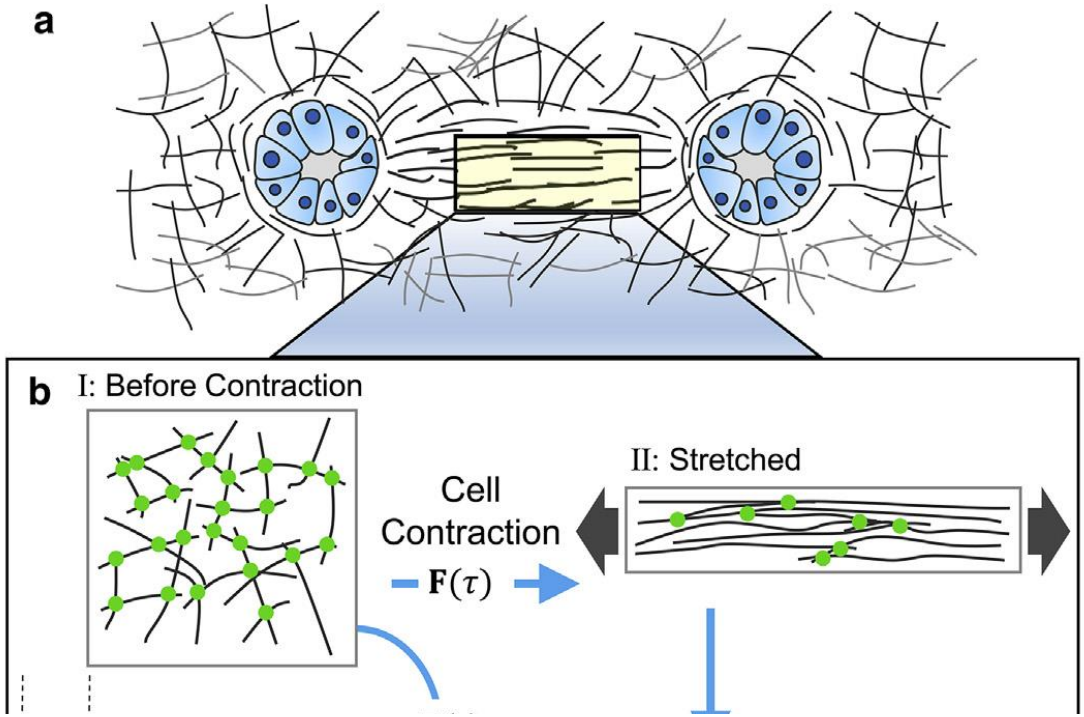
- Interactions of pairs of contractile cells in neo-Hookean and fibrous matrices
- Maximum principal strain.
- Lengths of red lines represent the magnitude of the maximum principle strain and their orientations show the directions of fiber alignment.



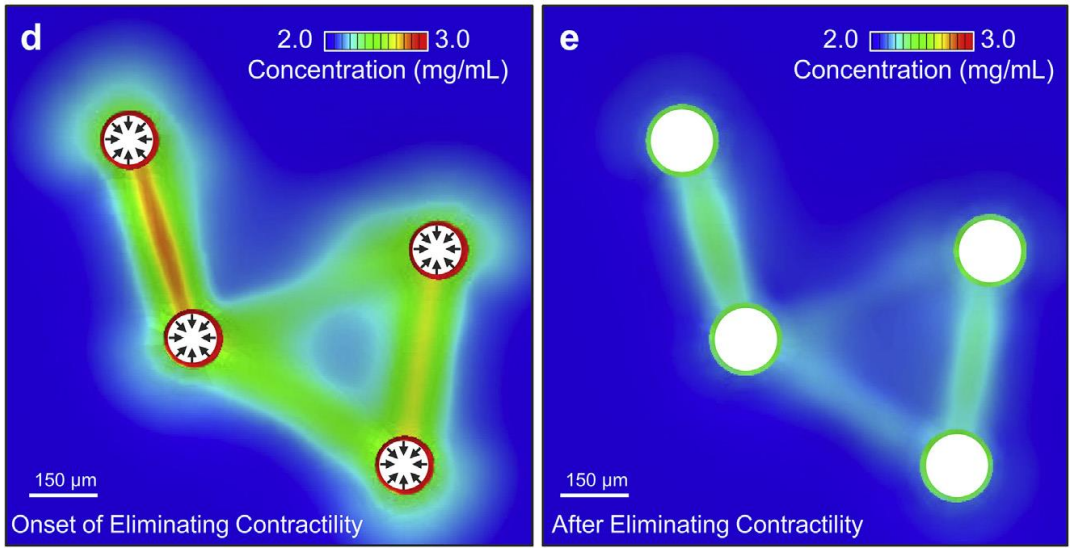
Plasticity

- Formation of weak crosslinks enhanced at increasing stretch ratio
- Association and dissociations rates

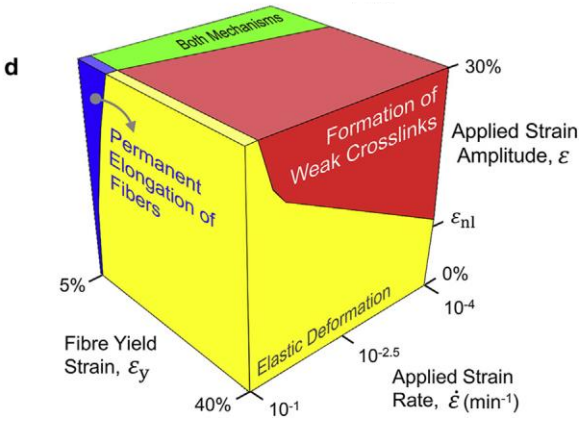
$$\bar{k}_{\text{off}} = \bar{k}^{0}_{\text{off}} \exp((\lambda_{\max} - 1)/\bar{\varepsilon}_0)$$



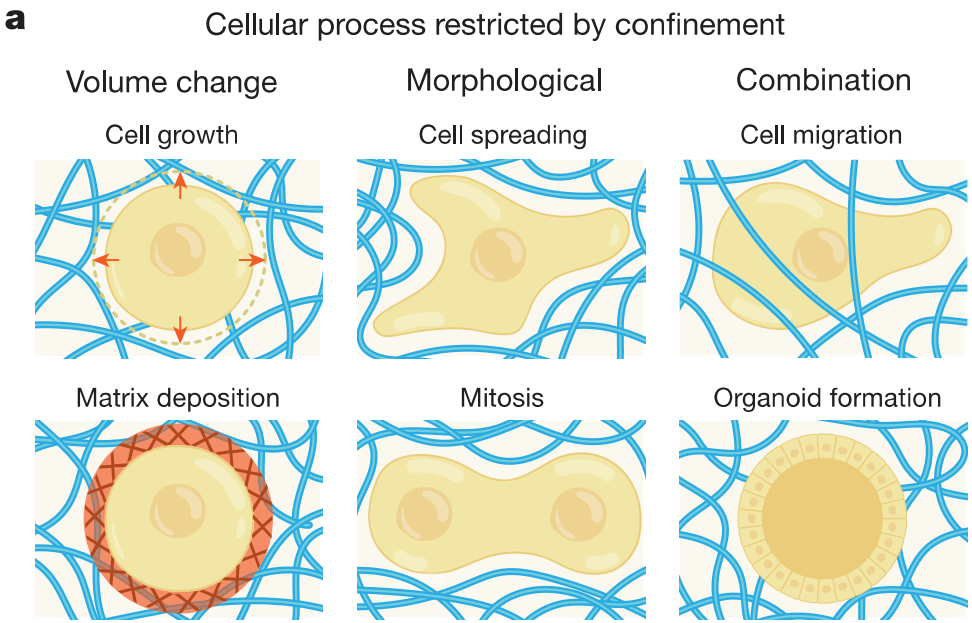
Plasticity

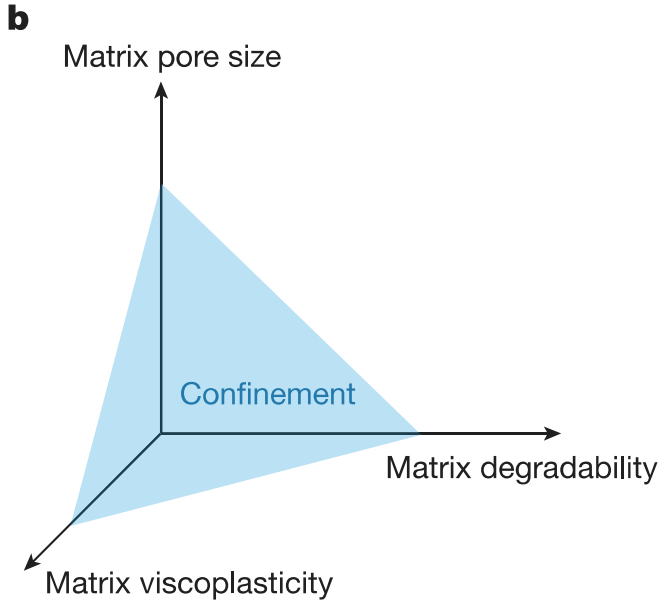


- Permanent elongation of fibers
- Strain rate and amplitude and mechanical properties of fibers

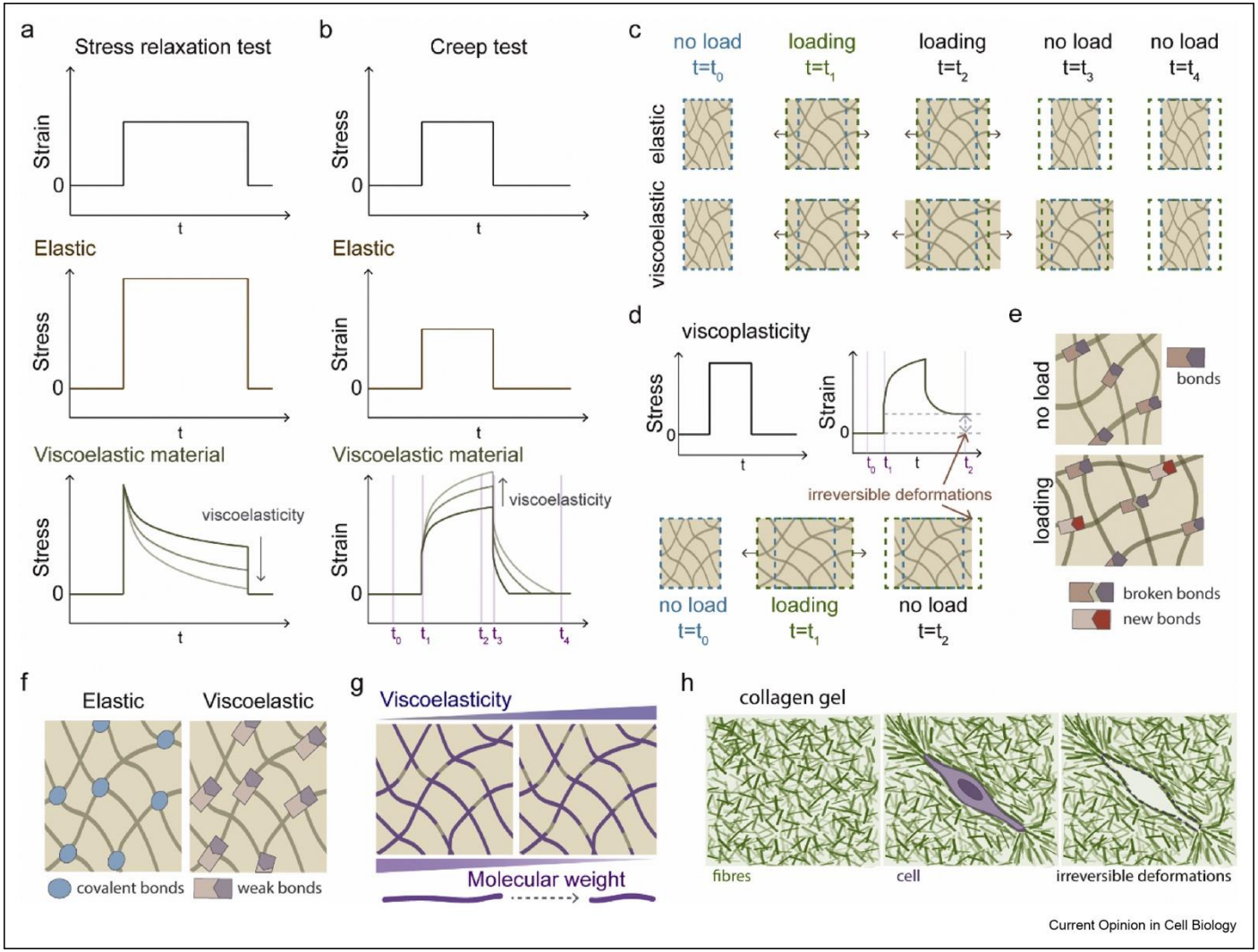


Matrix viscoplasticity



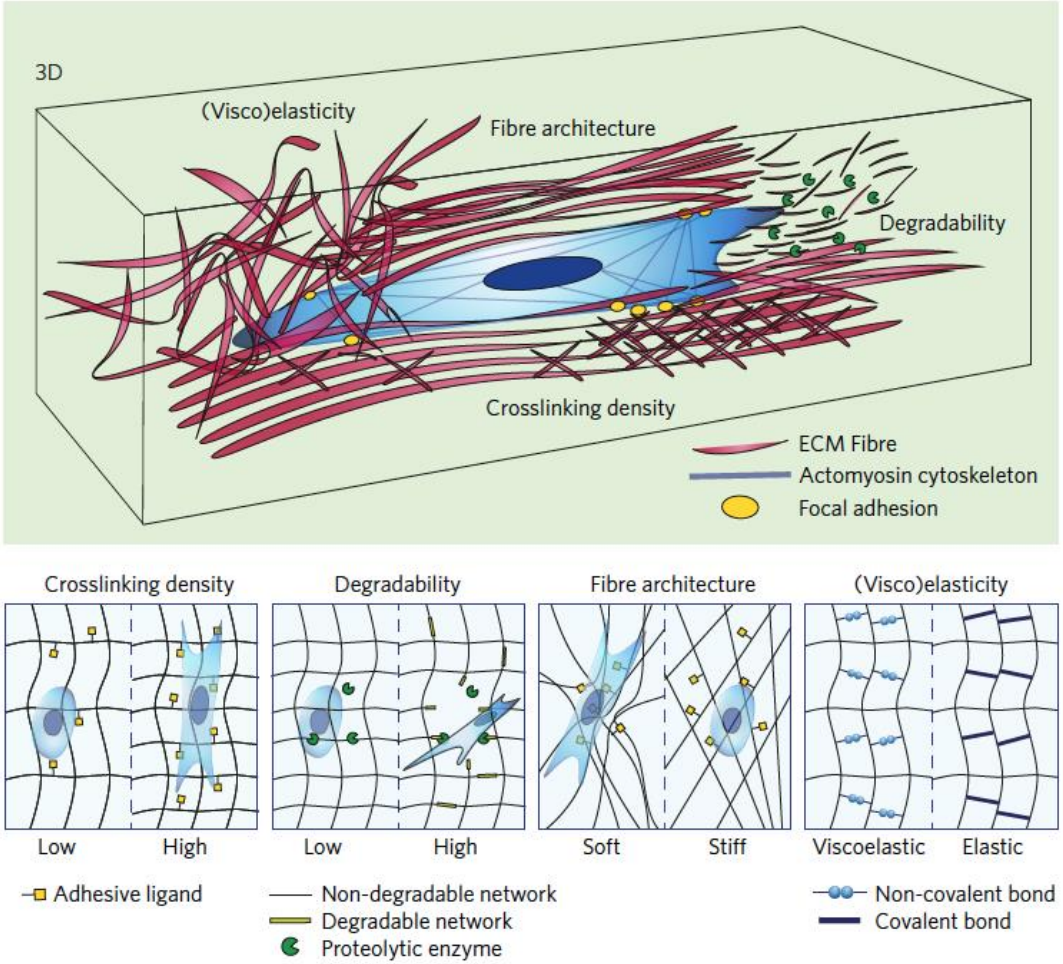


Matrix mechanics



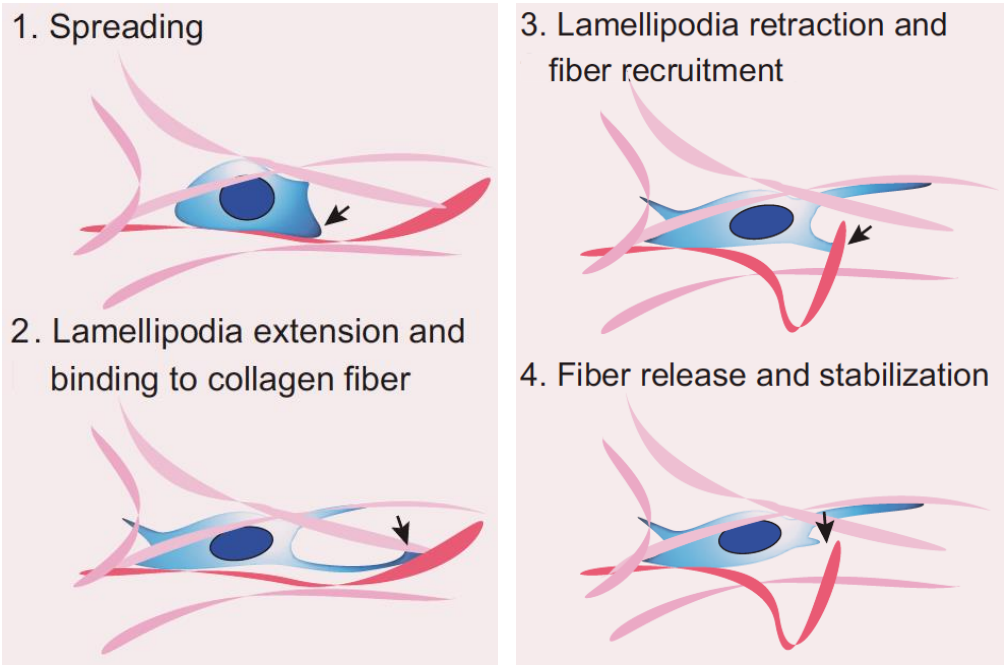
Engineering biomimetic biomaterials

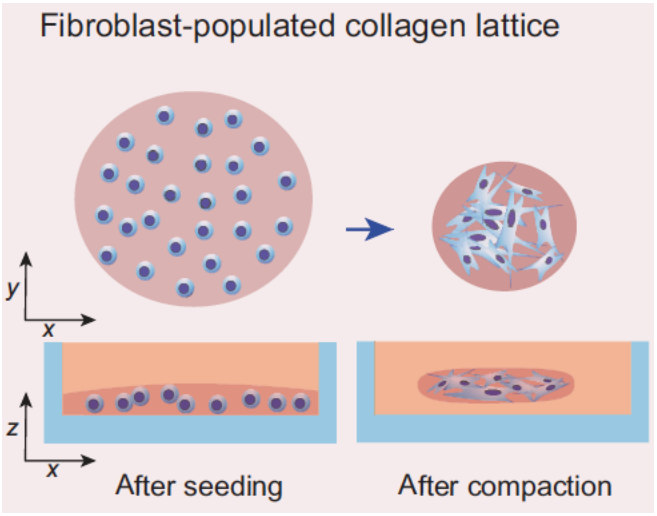
- Combinatorial effects of various factors

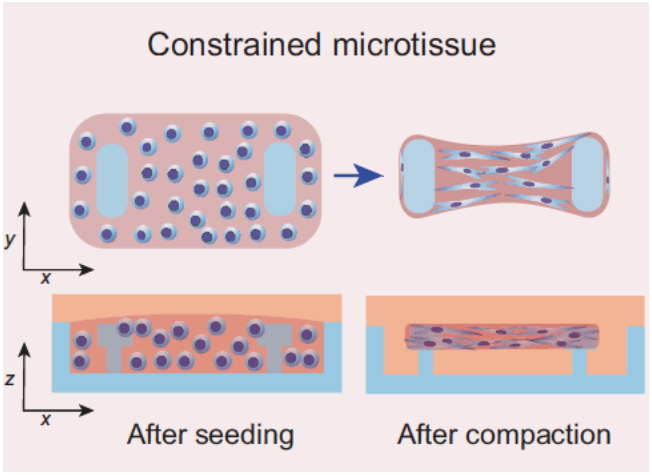


3D culture models of tissues under tension

- Cell-ECM compaction

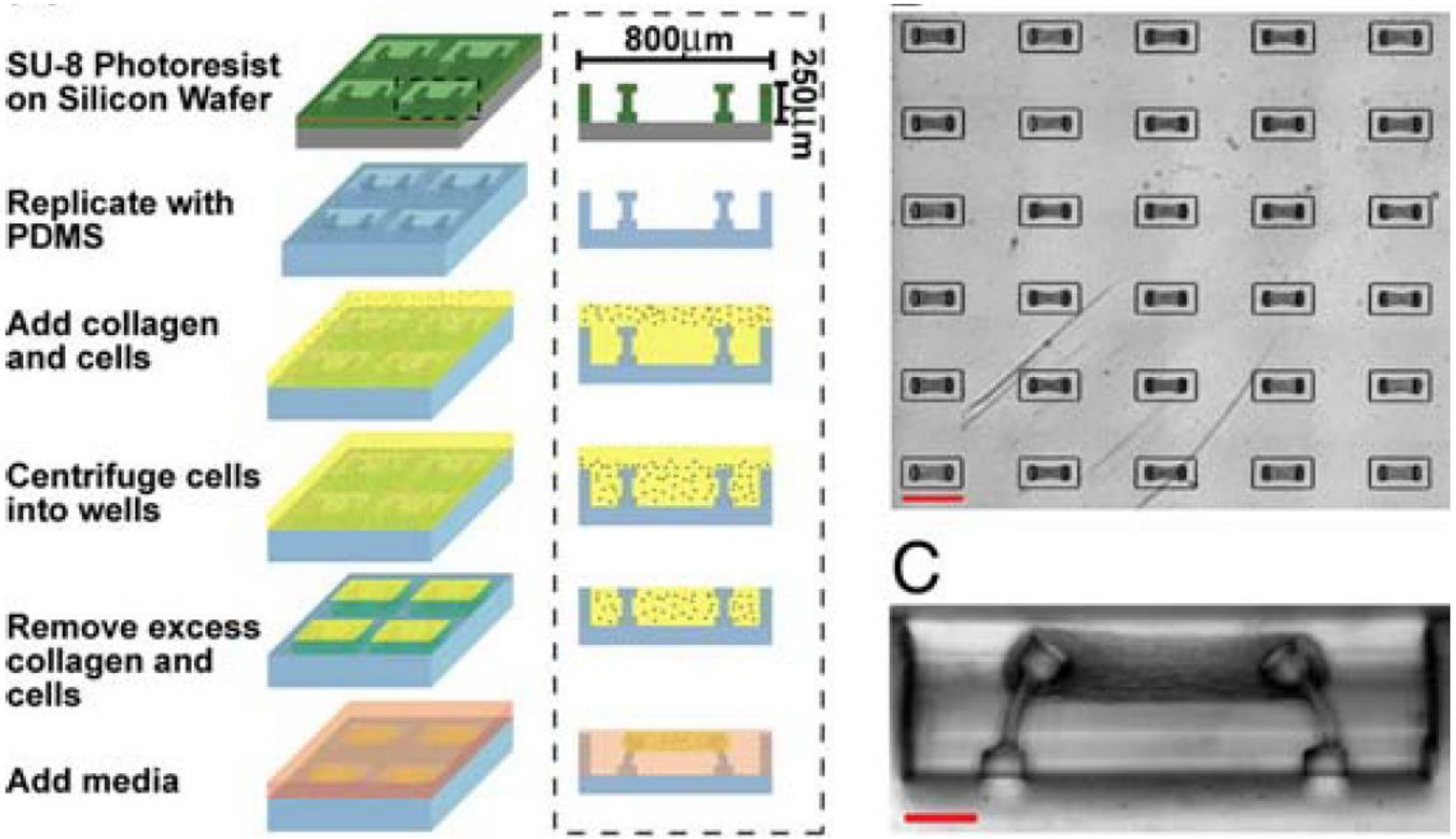




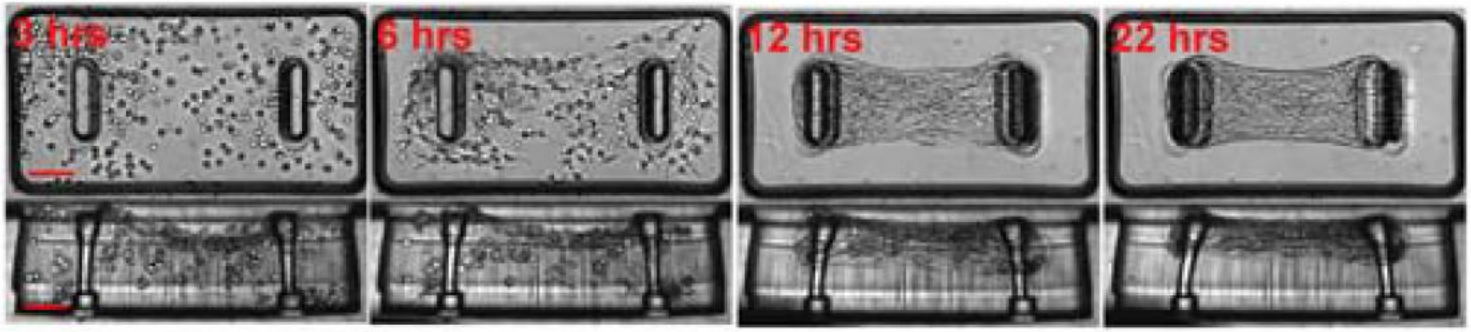


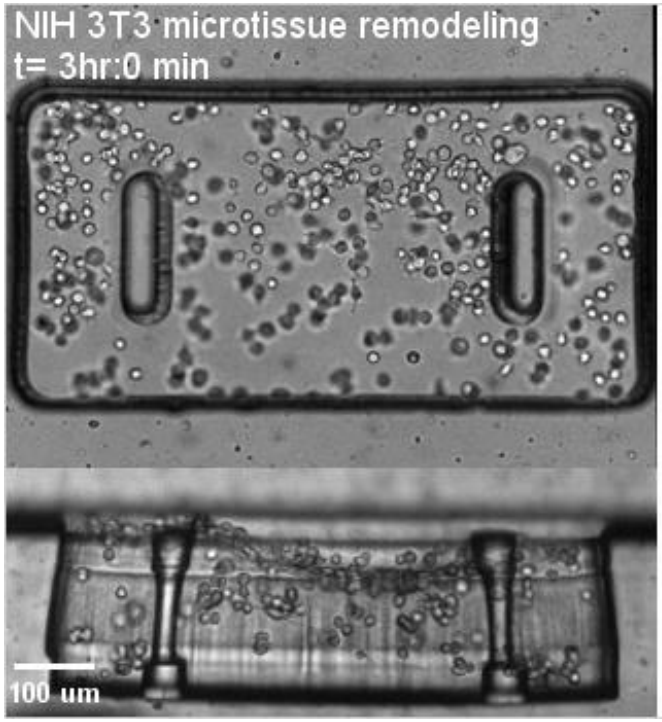
3D culture models of tissues under tension

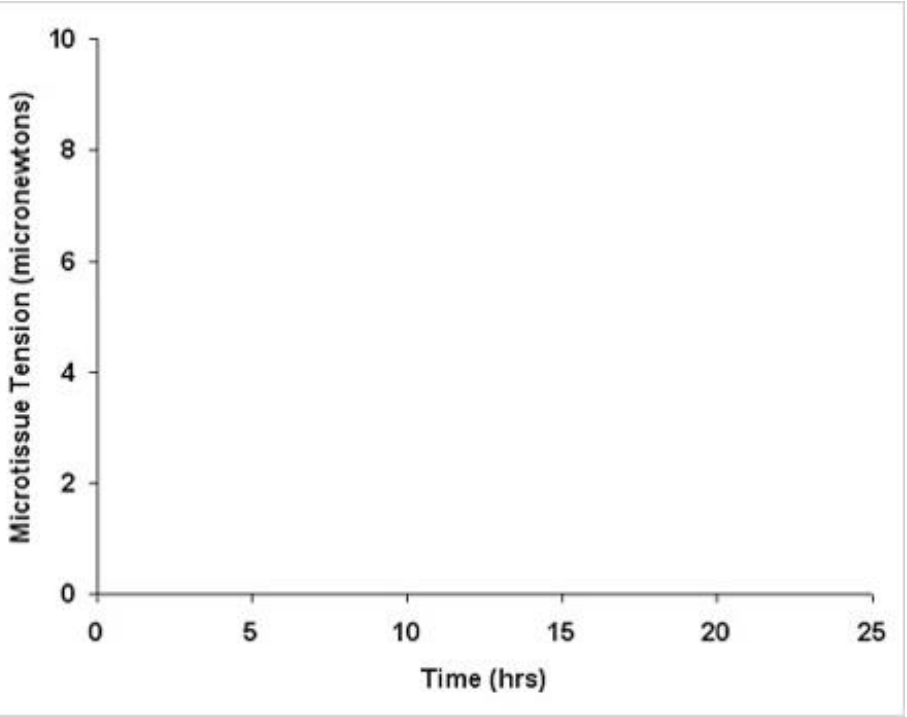
- Microfabricated tissue gauges



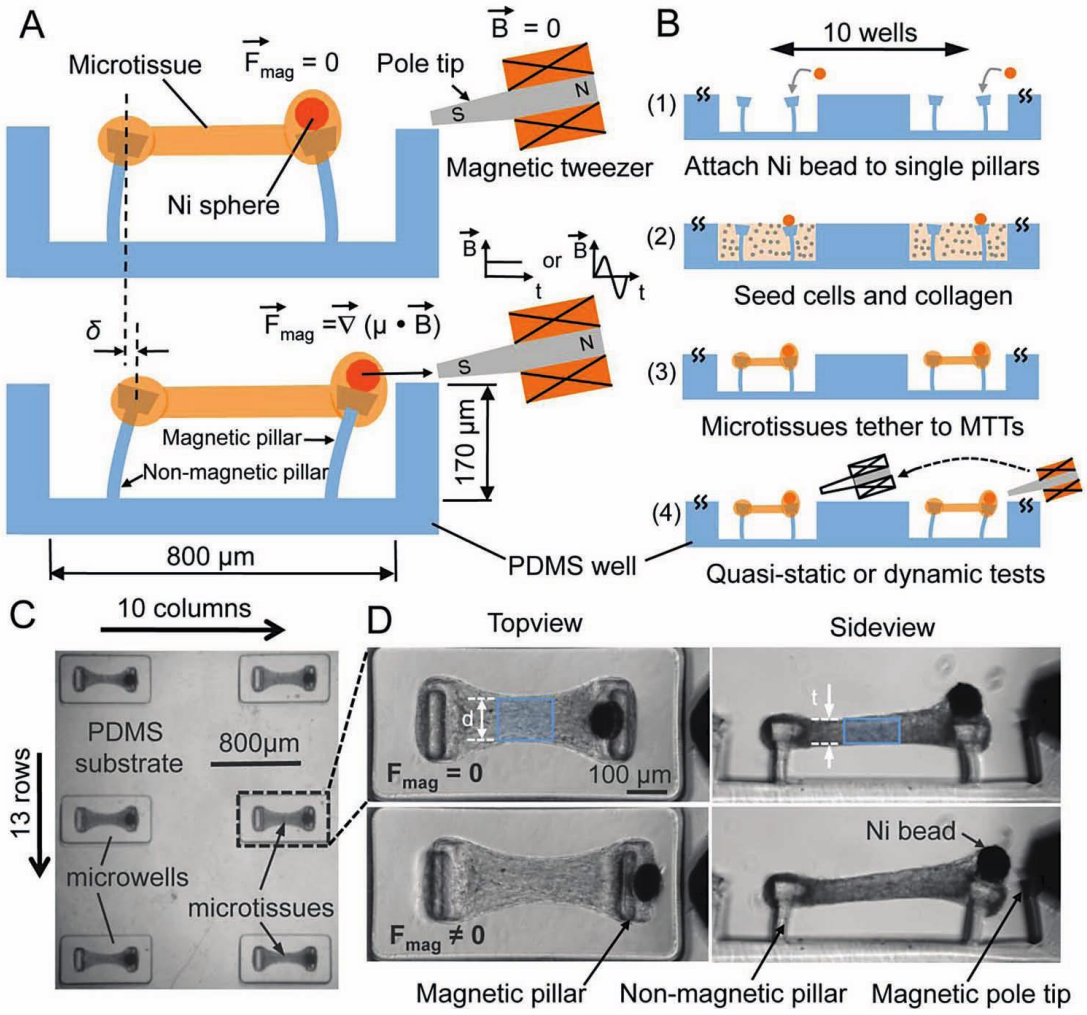
3D culture models of tissues under tension



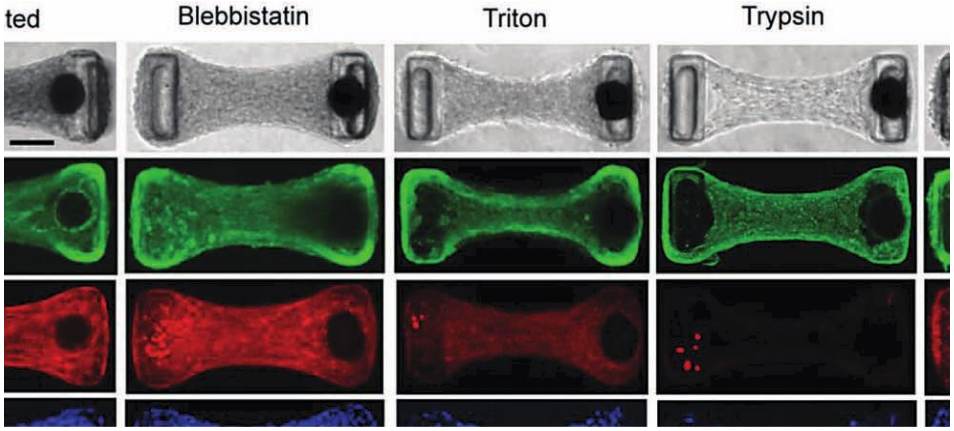


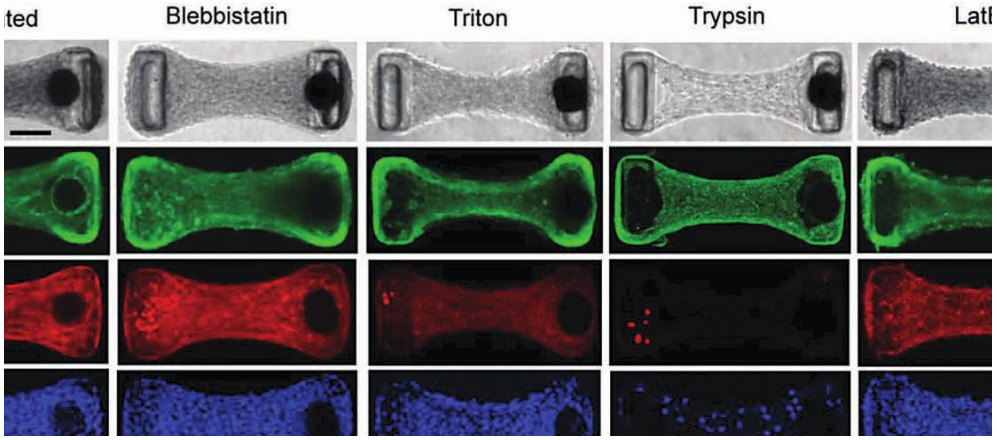


Mechanical characterization



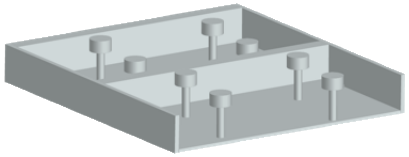
Mechanical characterization



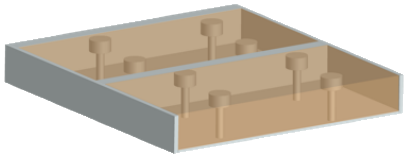


Contractile tissue mechanics: gap closure

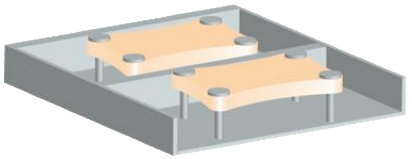
Microfabricated device

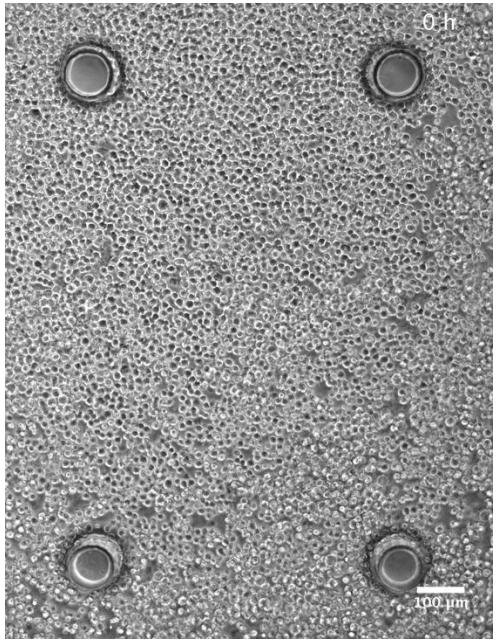


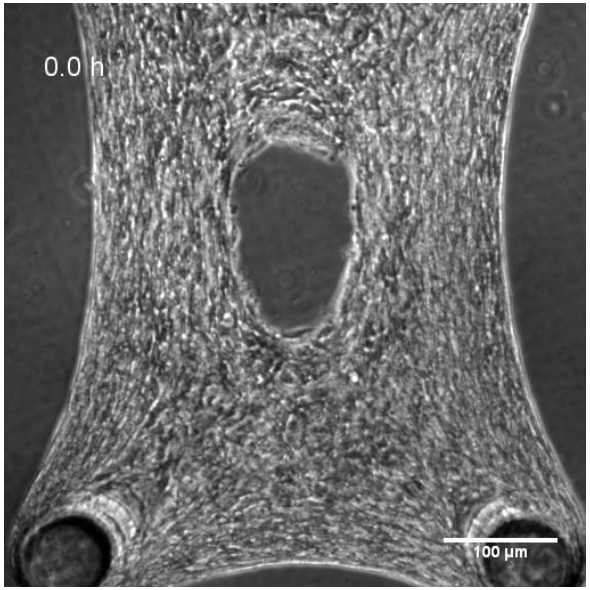
Cell/collagen suspension

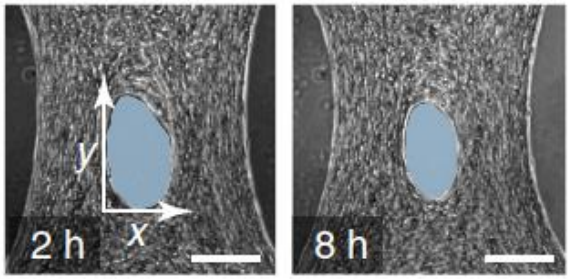


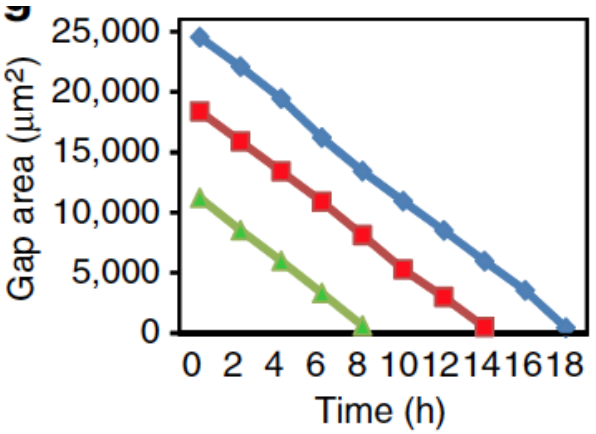
Tissue self-assembly





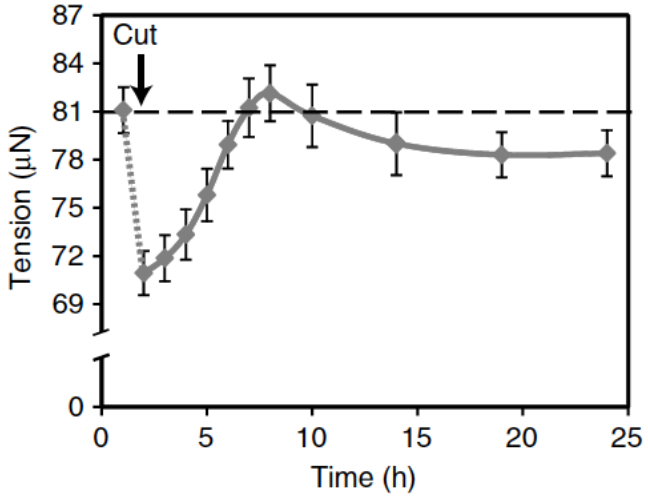


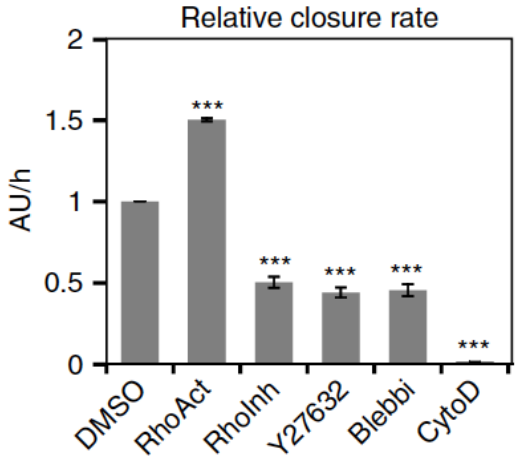


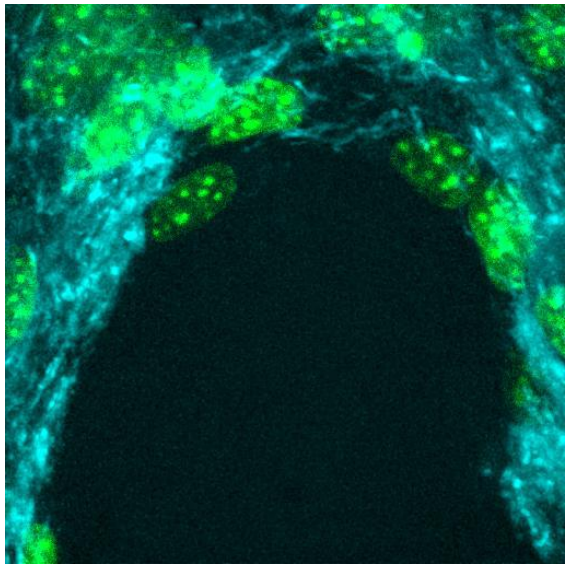


Contractile tissue mechanics: gap closure

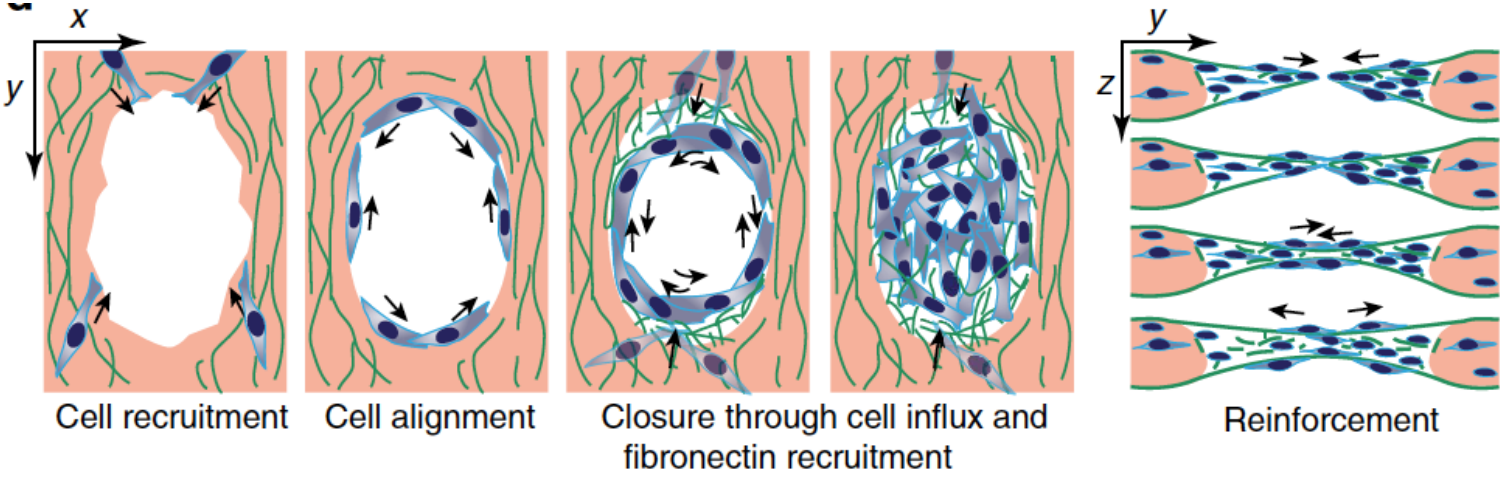
- Cell crawling? Purse-string?



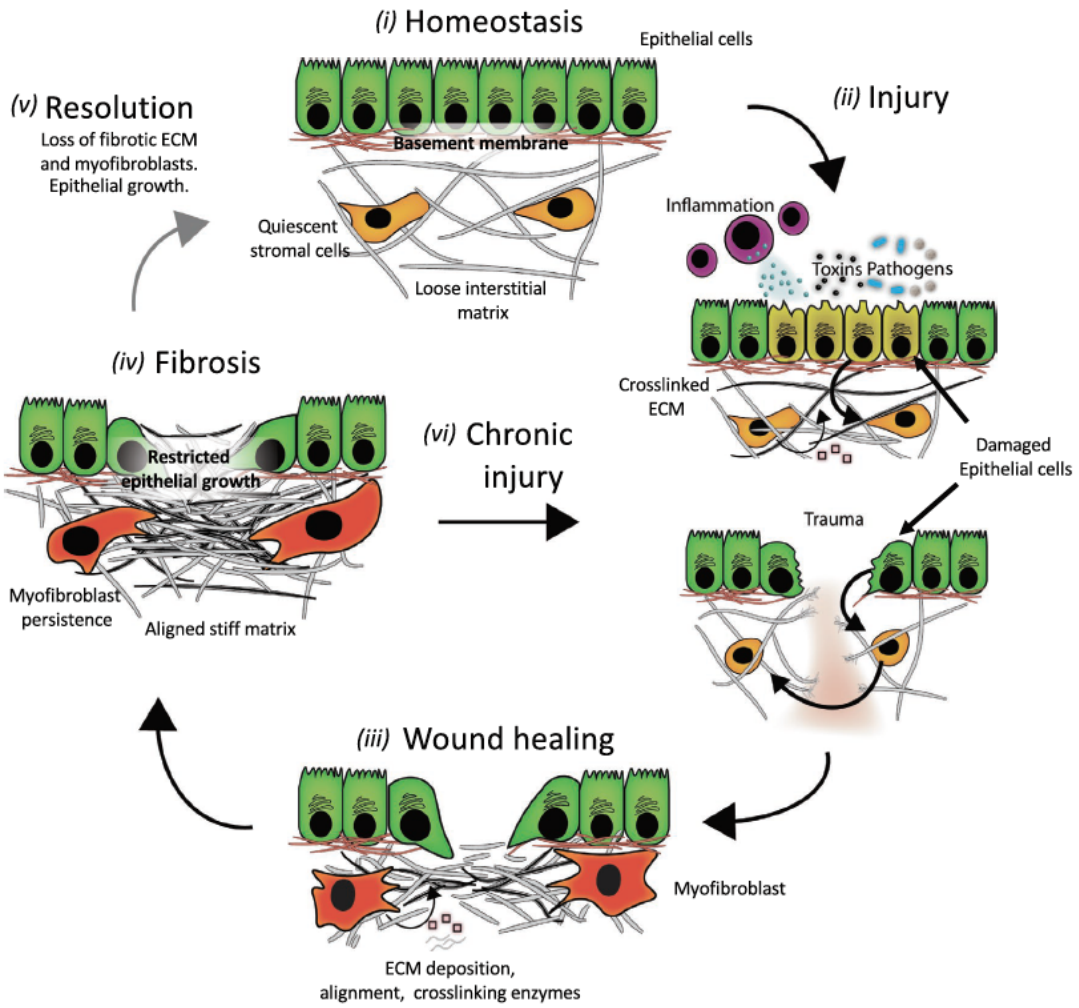




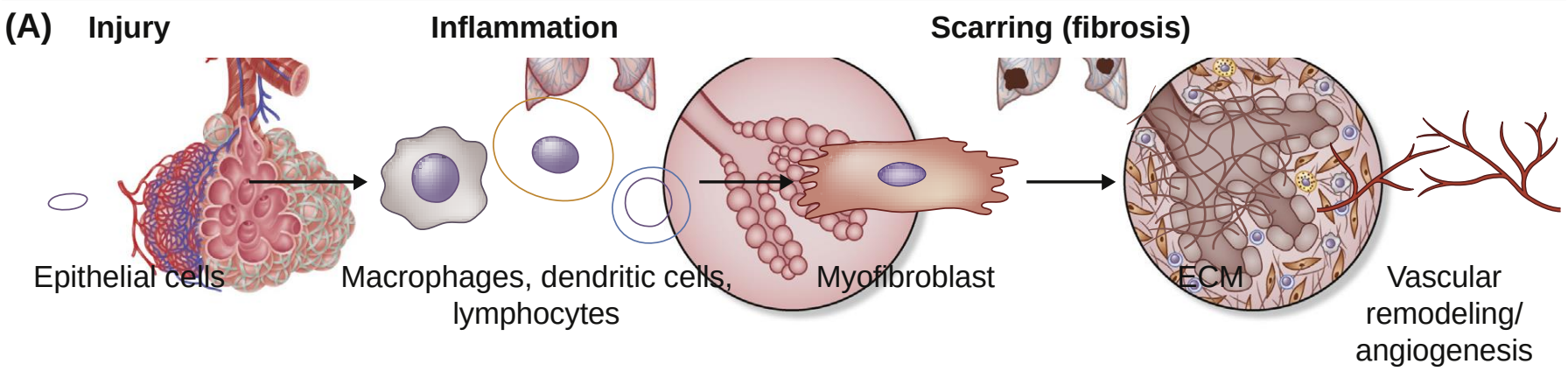
Fibronectin-488 / Nuclei



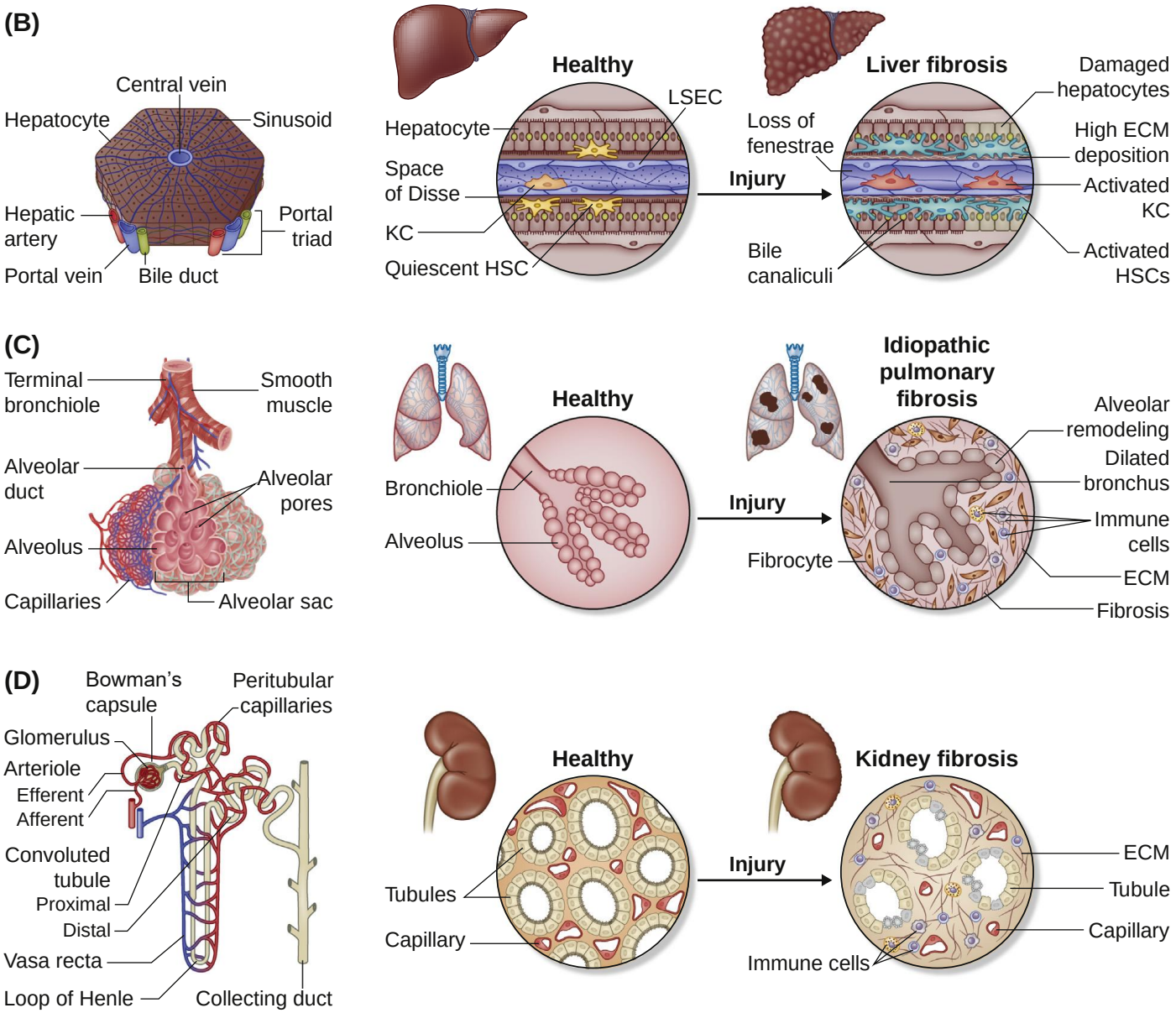
Tissue fibrosis



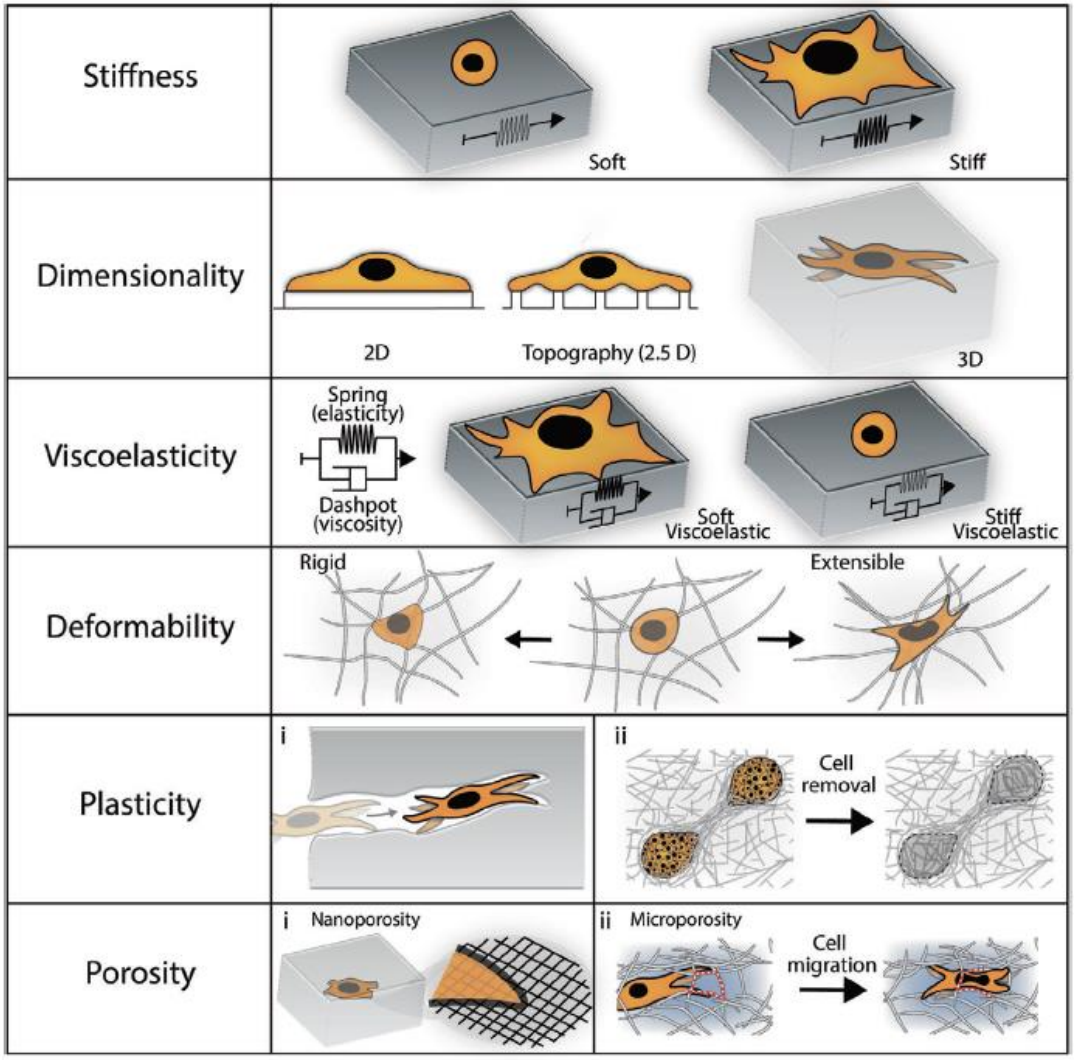
Tissue fibrosis



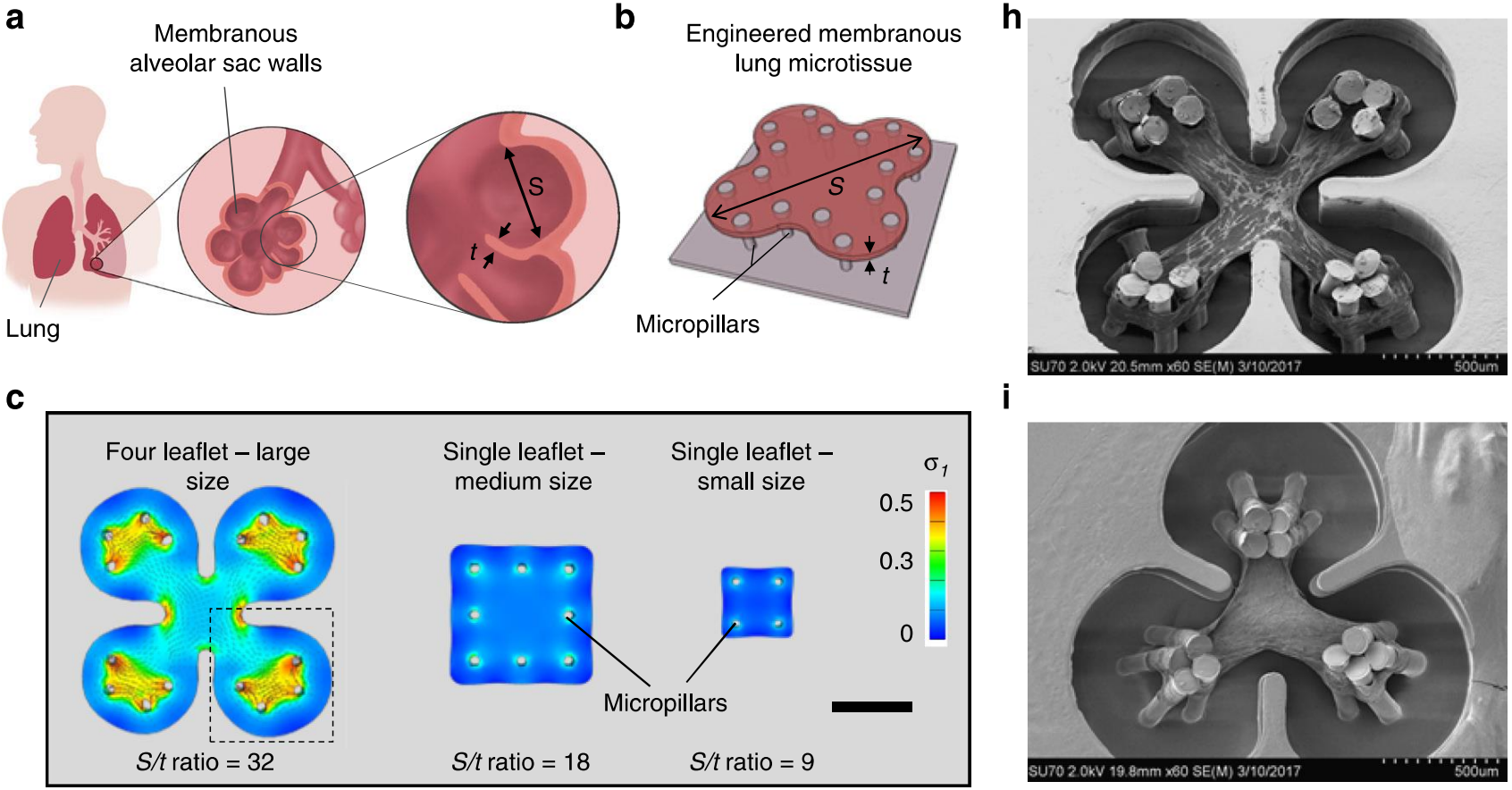
Tissue fibrosis



Platforms to study fibrosis



Platforms to study fibrosis



Platforms to study fibrosis